

TUTORIAL

A Model Qualification Method for Mechanistic Physiological QSP Models to Support Model-Informed Drug Development

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Mechanistic physiological modeling is a scientific method that combines available data with scientific knowledge and engineering approaches to facilitate better understanding of biological systems, improve decision-making, reduce risk, and increase efficiency in drug discovery and development. It is a type of quantitative systems pharmacology (QSP) approach that places drug-specific properties in the context of disease biology. This tutorial provides a broadly applicable model qualification method (MQM) to ensure that mechanistic physiological models are fit for their intended purposes.

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The use of modeling in drug discovery and development has received both industry and regulatory support in recent years.^{1–14} Some methods, such as pharmacokinetic (PK), population PK, and pharmacodynamic modeling, are now used routinely. Newer approaches, such as physiologically based PK, systems biology, and QSP, are being used effectively in a number of organizations, but are still in the early stages of broad market adoption.^{4,15,16}

QSP is an umbrella term for modeling approaches that integrate a mathematical representation of the biological system with pharmacological information about a drug of interest in order to facilitate improved understanding of human drug response. As summarized in an National Institutes of Health QSP White Paper, QSP is intended to help identify and validate targets, reveal possible biomarkers, support drug design, inform dose and regimen selection, and help identify (non-)responders proactively.¹⁰

Mechanistic physiological modeling is a QSP approach in which the mathematical representation of the biological system comprises known and hypothesized dynamic relationships between biological components that give rise to systems-level (e.g., clinical) behaviors. One or more drugs' effect(s) are then represented mechanistically in the context of the biological system. This approach facilitates improved understanding of the relationships between biological components (e.g., organs, cells, mediators, signaling pathways), the effect(s) of the intervention(s) (e.g., receptor agonism, transport inhibition), and outcomes of interest (e.g., plasma glucose levels, tumor size, markers of inflammation). Engineering principles are applied to translate biology into graphical and mathematical expressions. This translation process relies on both engineering and life science expertise. In terms of complexity, such models typically contain more mechanistic biological detail than mechanistic or semimechanistic PK/pharmacodynamic models (see refs. 17 and 18 for examples) but less pathway-level detail than systems biology models (e.g., see refs. 19 and 20). The biological entities modeled in mechanistic physiological

modeling often span across scales, from molecules to pathways to organs or whole organisms.^{21–23} Most commonly, the models are systems of ordinary differential equations, which are well-suited for representing systems with dynamic interactions between components.^{23–25} Unlike many other modeling approaches, mechanistic physiological modeling does not rely on any single type of data to infer model structure and parameterization. Rather, it is a scientific method that combines relevant available data with scientific knowledge and engineering approaches to construct plausible representations of biology in order to better understand the biological system. These attributes make it a natural fit for drug discovery and development, in which improved understanding of how modulation of a target affects clinical outcomes can greatly improve decision-making.

Pathophysiology and drug action are complex, and drug development typically requires making decisions under conditions of uncertainty, using implicit and explicit assumptions about the role of a drug or target in the disease process. Typical questions facing drug developers include:

- Will the target pathway show sufficient efficacy to be competitive?
- What is the likely human clinical efficacy taking into account systemic feedbacks and compensatory mechanisms?
- How much of a risk does biological uncertainty pose?
- What aspects of biology must be clarified before moving forward and what are the most informative experiments?
- Could patients with a range of disease severities benefit from this drug?
- Are there more and less responsive patient subsets and, if so, how can they be identified?
- Which compound properties should be optimized?
- Might combination therapy be more promising than monotherapy?

Decisions about the next steps in drug development are typically made with limited or no clinical data and no clear answers to all of these questions. Further, the typical drug development process is not designed to answer all of the

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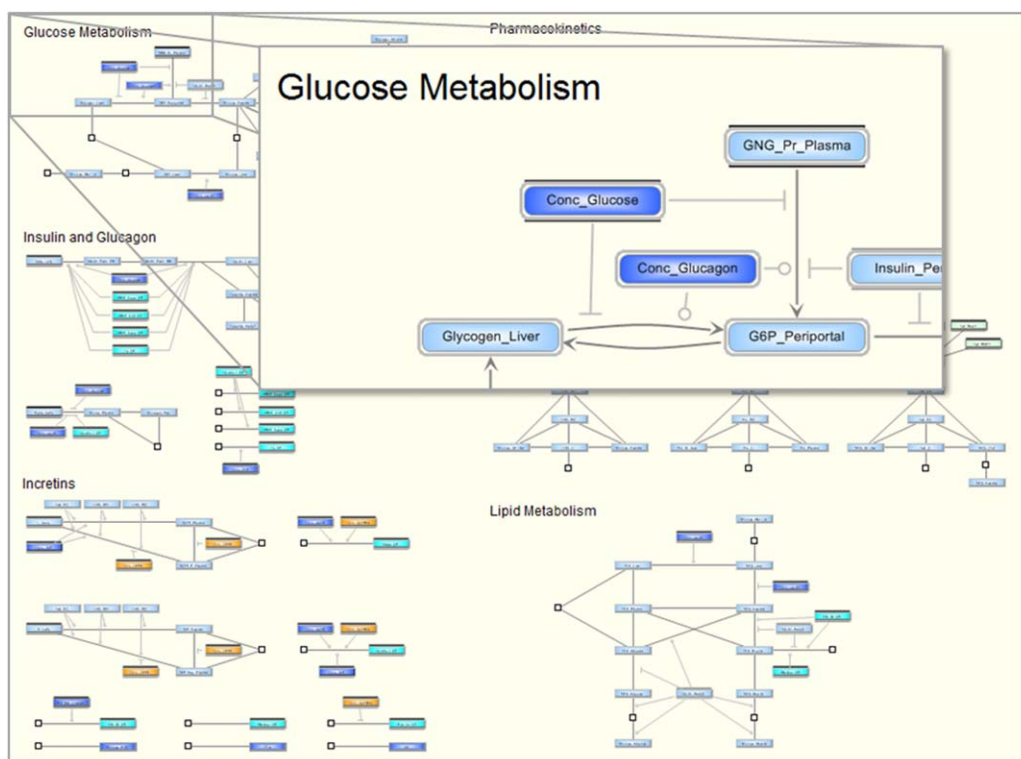


Figure 1 A portion of a graphical representation (PhysioMap) of a type 2 Diabetes PhysioPD Research Platform developed by Rosa & Co using JDesigner software.⁸⁶ Major biological processes are represented graphically as subsystems or modules (e.g., glucose metabolism, insulin and glucagon, incretins, etc.). They are linked mathematically by the use of an aliasing function that allows display of the same node in multiple places on the diagram. The detailed section represents key regulated processes in glucose metabolism. For example, glycogenolysis (the reaction arrow going from liver glycogen ["Glycogen_Liver"] to glucose 6 phosphate ["G6P_Periportal"]) is regulated by glucose ("Conc_Glucose") and glucagon concentrations ("Conc_Glucagon"). The graphical layout facilitates communication and review of the model by research team members with biological content knowledge.

questions. Relevant pieces of information to help answer the questions may already exist, but often have not been connected formally. For example, to understand how an insulin secretagogue will affect plasma glucose concentrations, researchers must consider its PK properties, its concentration-dependent effect on insulin secretion and possibly other pathways, the role of insulin in regulating glucose, and feedbacks that could amplify or dampen response.^{24–26} Mechanistic physiological models, such as the example of a type 2 Diabetes PhysioPD Research Platform developed by Rosa & Co (**Figure 1**), have been used to integrate relevant existing information in order to address these types of questions explicitly.

The evidence informing the model representation may include *in vitro* assay results, preclinical animal data, PK data, clinical data for the compound of interest or for drugs with related mechanisms of action, as well as the larger body of work establishing hypotheses of disease pathophysiology. This information already informs the mental models in scientists' heads, but human brains are not equipped to fully integrate and utilize all of the information.^{27–31} Mechanistic physiological modeling makes this integration task explicit, consistent, and testable, making use of relevant available data to optimize decisions throughout the drug development process. As more data become

available, the model continues to evolve to help anticipate the next development step with the most complete understanding possible, very much in keeping with a learn and confirm approach.³² The goal of this type of modeling is to empower scientific dialog, not to replace it.

Mechanistic physiological modeling has been utilized since well before the term QSP came into general use. For example, in a 2005 publication, Rullmann *et al.*³³ demonstrated the use of a mechanistic rheumatoid arthritis model for the identification and prioritization of novel targets based on improved mechanistic understanding of disease dynamics and prediction of likely clinical efficacy. In 2007, Gadkar *et al.*³⁴ demonstrated the use of a type 1 diabetes model for explaining apparently contradictory data and using novel insights to design optimal protocols. At this date, there is a record of publications and conference presentations in a wide variety of therapeutic areas, including type 2 diabetes,^{21,35–40} type 1 diabetes,^{34,41,42} rheumatoid arthritis,^{33,43–45} skin diseases and sensitization,^{46–49} liver toxicity,^{50–54} asthma,^{55,56} bone remodeling,⁵⁷ cardiovascular disease,^{44,58,59} and oncology/immuno-oncology.^{60–62} Published applications of mechanistic physiological models include clarification of biological mechanisms and hypothesis exploration,^{40,41,46,48,50,52,54,55,57–59,61} target identification and evaluation,^{33,48,49,58,63} compound evaluation or

Table 1 Aspects of model qualification and example questions that model qualification must address

Resolution	Conditions under which the resolution is appropriate
Relevance	<ul style="list-style-type: none"> • Is the research context clear and has biological and functional scope been set accordingly? • Does the representation of the biology and pharmacology reflect the current state of knowledge?
Uncertainty	<ul style="list-style-type: none"> • Given biological uncertainty, how robust are model results and conclusions? • Would conclusions or recommendations change under different assumptions? • What uncertainty poses a risk to the program, and what experiments could resolve it?
Variability	<ul style="list-style-type: none"> • How do known differences between patients affect model results? • What biological mechanisms can explain the range of observed clinical or preclinical outcomes?
Data	<ul style="list-style-type: none"> • Does the model match relevant data, at the clinical/preclinical and mechanistic level? • Are model responses to a variety of tests consistent with current knowledge and expectations?

optimization,^{39,45} preclinical to clinical translation,^{45,51,59} experimental design and protocol optimization,^{34,38,42,53,60,62,64} patient stratification,^{45,62,64,65} and biomarker identification.^{43,46}

Most of these examples illustrate uses of mechanistic physiological modeling to inform internal drug development decisions. Notably, a well-established QSP model of bone and calcium homeostasis was also recently utilized by the US Food and Drug Administration to inform a regulatory decision.⁶⁶

THE NEED FOR A MODEL QUALIFICATION METHOD

Despite a number of successful applications and increased investment and support for the use of modeling in pharmaceutical companies,^{1–3,5,12–14,16,21,23–25,33,41,66} many decisions made in pharmaceutical discovery and development today still do not fully benefit from systematic and targeted application of biological modeling and simulation. One possible reason for the relative underutilization of QSP approaches, such as mechanistic physiological modeling, is that the field is lacking a standard model qualification approach that would give decision-makers confidence in assessing model quality and acting on model-informed insights. Such a qualification approach could also help establish a reasonable standard for reviewing models for publication. Finally, one of the necessary conditions for using a QSP effectively is an organizational structure and culture that enables cross-functional communication. A standard qualification approach would facilitate communication between modelers and other members of cross-functional teams whose scientific expertise is essential for model development and qualification, but who may be unsure how to assess model quality.

The purpose of model qualification is to ensure that a model is fit for the purpose for which it is intended. Before determining how to qualify mechanistic physiological models, it is therefore

important to understand that the ultimate purpose of these models is to support decision-making in drug discovery and development. Decisions in drug discovery and development usually have to be made with incomplete data, especially in early development. A well-qualified mechanistic physiological model should make optimal use of incomplete but informative data and knowledge to de-risk the next steps in development. It is a common misconception that the goal of this type of modeling is limited to prediction of outcomes. Indeed, precise prediction is not always possible, but mechanistic modeling can still be used to clarify biological connections, explore the impact of different hypotheses, identify the factors most likely to affect outcomes, and suggest experiments to reduce material uncertainties. For example, a mechanistic physiological model can be used to explore best-case and worst-case scenarios and identify what would predispose a patient toward one or the other scenario. Important goals of mechanistic physiological modeling therefore include generation of new biological insights and testable hypotheses.^{24,28,29,68} A qualification approach to determine if such a model is “fit for purpose” must be designed with these purposes in mind. Early attempts at establishing model “validation”^{27,69} have missed this important point by focusing exclusively on predictive ability. This type of validation approach is neither necessary nor sufficient for these types of models. More recent publications have acknowledged that there are qualitative differences between QSP models and other modeling approaches that necessitate a fresh perspective on model qualification.^{4,17,70,71}

If traditional validation approaches cannot ensure that a mechanistic physiological model effectively serves the functions outlined above, then what approach is appropriate? We introduce here a broadly applicable model qualification method (MQM) designed to ensure that mechanistic physiological models are fit for purpose. This approach has been developed and proven effective in dozens of professional applications in a broad range of therapeutic areas.^{38,40,45,48,49,59,61,62,64,71} The criteria address four aspects of model qualification: (1) relevance to the research context; (2) dealing with uncertainty; (3) dealing with variability; and (4) ensuring that model results are qualitatively and quantitatively consistent with test data. **Table 1** lists some of the questions that need to be addressed for each of these aspects of model qualification. Use of the MQM brings structure to model-based research, creates a common language, and improves communication among and buy-in from cross-functional teams faced with making decisions at every step of the drug development process.^{4,71}

THE MODEL QUALIFICATION METHOD

The MQM presented here evolved specifically to support decision-making under the conditions commonly faced in drug development: incomplete information and limited time. Hence, the goal is to build the best and most useful model possible under these conditions. Of primary importance is to get buy-in from the development team that the model is fit for its intended purpose of supporting drug development research. **Figure 2** is a graphical summary of the MQM. We propose that these criteria are necessary and sufficient for model qualification.



Figure 2 Graphical illustration of the eight criteria of the model qualification method.

All criteria of the MQM must be considered in every modeling project, to the extent appropriate to the research context (defined in the section below).

The following sections discuss each of the criteria of the MQM and suggested qualification approaches to determine whether the criteria have been met. It should be noted that the MQM is a flexible framework rather than a strictly prescriptive method. That is, it is not a rigid set of steps that must be followed to satisfy each criterion. Such an approach would be too inflexible given the broad range of therapeutic areas and research questions that these models can help address and the wide range of data availability and uncertainty that may be encountered. Rather, the specifics of the qualification approach for each of the criteria in the MQM should be planned during the first stages of a modeling project. The purpose of the MQM is to ensure that all of the criteria that are relevant for model qualification are considered and documented from the initiation of the project.

As discussed above, two attributes that clearly distinguish QSP models from empirical approaches are the purpose of the modeling exercise and the data used for modeling. These attributes also vary between QSP models. The purpose of QSP modeling could range from target evaluation to phase III protocol optimization. The data available may be overwhelmingly mechanistic and preclinical, or could include a substantial amount of clinical data. It is important to tailor the model, the qualification approach, and indeed the team's expectations according to the intended application of the model and the available data. For example, an early evaluation to support the biological case for advancing a novel target can often be made using primarily preclinical mechanistic data and the broader disease literature. A fit-for-purpose model in this scenario may not yet need to account for possible mechanistic

drivers of clinical variability, although it should consider the impact of biological uncertainty on the go/no-go decision for the target. On the other hand, a QSP model used for phase III protocol optimization should include a range of virtual patients to account for clinical variability and should certainly be tested against clinical data from earlier phases (i.e., the model qualification criteria must fit the data and intended use of the model).

There is active ongoing research on techniques to address many of the areas discussed below (see refs. 72–79 for examples). The focus here is not on specific techniques but on: (1) elucidating the need for each of the criteria; (2) suggesting qualification strategies; and (3) bringing all of the criteria together into a complete framework to appraise a model's fitness for purpose.

MODEL SCOPE IS RELEVANT FOR RESEARCH CONTEXT

The first criterion requires that the model scope be relevant for the research context. A costly and unfortunately common problem in modeling-based research is building an inappropriate model for the research context. For example, models may have more detail than needed in areas that are not of primary relevance for the current research question, which could contribute to failure to address the key questions in time to inform decisions. We submit that the research context comprises: (1) the key research question(s) or decision(s) to be made; (2) the available data and knowledge; and (3) time and resource constraints.

Model scope should be parsimonious and based on relevance to the research context.^{4,70,71} The determination of which biological mechanisms to include in a mechanistic physiological model should primarily be based not on what is known or what data are available, but rather on what is most relevant to the research context and what is sufficient to answer the research question. For example, in building a model to explore mechanistic explanations of the two-phase bronchodilatory effects of zileuton in patients with asthma, Demin *et al.*⁵⁵ focused only on key drivers of bronchodilation and cellular dynamics that were relevant to the timeframe of the clinical observations.

This qualification criterion should be the foremost consideration when planning the project and designing the model. Every scope decision must be made with the research context in mind. Given time and resource constraints, inclusion of extraneous detail can be as detrimental as exclusion of needed components.

Recommended qualification approach

Begin the model-based research project by clarifying the research context. Before, during, and after the project, ask the following questions. If they can be answered in the affirmative, then the model was scoped appropriately.

- a. Did the modeling work yield actionable insights that advanced the research agenda?

Examples of actionable insights include recommendations for optimal protocols, targeted follow-up experiments to de-risk next steps,

potential biomarkers predictive of efficacy or safety, or guidance for follow-on compound properties.

- b. Was the model constructed in a timely fashion so as to be useful when decisions had to be made?

It is critical to keep the available timeframe in mind when scoping models. It is often possible to stage the modeling work such that early insights can be gained in time for the most proximal decision, and additional functionality is then added in a staged fashion to support ongoing development. Models should have as much detail as is necessary for the research context, and no more. Unnecessary detail can delay or obscure insights into how mechanisms relate to outcomes and make it more difficult to augment or modify the model for future applications.

MODEL REPRESENTS RELEVANT BIOLOGICAL MECHANISMS

Mechanistic physiological models must represent relevant biological mechanisms at the level of detail appropriate to the research context.⁷¹ It is not uncommon for data to be most limited in precisely those areas that are of greatest interest to explore. Building a mechanistic physiological model in this situation is a way to integrate relevant information and knowledge into one framework that can then be used to test hypotheses and evaluate uncertainties in a systematic fashion (more on this in sections below). Mechanistic physiological modeling is particularly valuable when specific target or compound data are sparse because these models facilitate the representation of target or compound information and hypotheses within the context of relevant biological mechanisms. For example, Moore *et al.*⁶⁵ were able to use a mechanistic model of erythropoietin synthesis, binding, degradation, and cell dynamics to extrapolate from a single-dose study of a drug compound to assess likely hemoglobin response to a multidose protocol in a range of patients with kidney disease. This extrapolation was possible because the systemic effects of the drug and feedbacks that affect repeat-dosing PK were largely because of biological mechanisms represented in the model that were independent of the particular properties of the compound under investigation.

The requirement that mechanistic physiological models represent biological mechanisms imposes an important and rich set of constraints on parameters, equations, and subsystem behaviors. Parameters with physiological meaning can be derived using various types of data. Functional equation forms are chosen to be appropriate for the interaction that is represented (e.g., receptor binding), and subsystem behaviors can be compared to a rich set of experiments elucidating mechanisms (more on comparison to data in sections below). These attributes are particularly important in supporting simulations for novel drugs or protocols (e.g., for planning an optimally informative phase I trial). Note that criterion 2 does not imply that all known biological mechanisms must be represented, as that would violate criterion 1. Similarly, the level of mechanistic detail represented should be informed by the research context. For example, if the compound under investigation is a receptor agonist, the details

of receptor binding and receptor recycling dynamics may be very relevant to include, but the interactions of other ligands and receptors in the same model need not have the same level of mechanistic representation.

Recommended qualification approach

Document literature information, data, and the scientific judgments and data interpretations that inform the model structure, equations, and parameters. All parameters must have biologically reasonable values. In addition, it is imperative that scientific experts on the team contribute to the modeling process, understand the model structure, and confirm that the represented interactions between components reflect the biology appropriately. (Disagreement among experts suggests uncertainty, which can be addressed as discussed in sections below.) To facilitate communication within a multidisciplinary team, it is extraordinarily helpful to use modeling software with a graphical interface and annotation capabilities.

RELEVANT QUALITATIVE UNCERTAINTIES ARE ASSESSED

Inevitably, explicit modeling of a biological system leads to identification of gaps in current knowledge. Knowledge gaps and uncertainty exist irrespective of modeling; constructing a model brings much-needed rigor to the identification and assessment of uncertainty so that the best possible decisions can be made in the presence of uncertainty. Typically, only a small subset of uncertainties significantly affect outcomes, and identification of these uncertainties can help determine the most informative experiments or approaches to resolve them.

Assessment of the possible impact of mechanistic uncertainty is a particular strength of mechanistic physiological modeling. These models lend themselves exceptionally well to scientifically motivated explorations of “what-if” scenarios that do not necessarily require specific data or assessed distributions *a priori*. The motivating questions may be, what if patients’ pathophysiology varied in one or two pathways, would this pose a potential risk (i.e., some types of patients would respond poorly) or would it present a potential opportunity (i.e., some patients would respond particularly well)? Or, if we do not know which of several mechanistic hypotheses is correct, how much might that affect outcomes?

In the MQM, we distinguish qualitative and quantitative uncertainty. Qualitative uncertainty is uncertainty about how biological components interact (e.g., should there be an interaction between necrosis and activation of antigen presenting cells? What is the functional form of the equation?), whereas quantitative uncertainty is uncertainty about the degree or rate of the interaction (e.g., to what extent does interleukin-10 inhibit T-cell activation in the presence of other mediators?). Uncertainty about the identity of the compound’s molecular target is an example of a qualitative uncertainty, whereas uncertainty about the degree to which target modulation will impact downstream functions is a quantitative uncertainty.

To assess a qualitative uncertainty means to evaluate its impact on the research question. Often, closer examination of an uncertainty leads to resolution, where some hypotheses can be ruled out as unlikely given all the existing

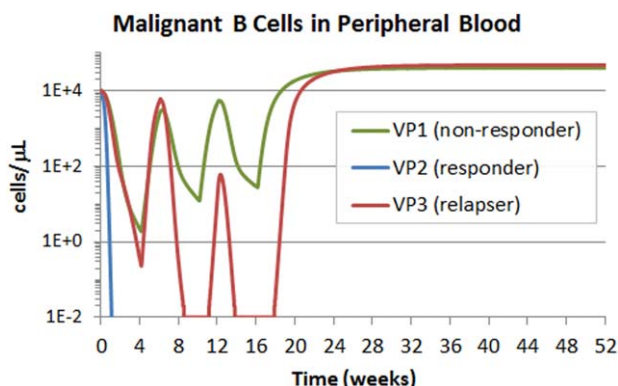


Figure 3 Illustration of the virtual patient (VP) concept. Several VPs were created to explore hypotheses of patient differences underlying response or nonresponse to three cycles of blinatumomab, a bi-specific T-cell engaging antibody in B-lineage acute lymphoblastic leukemia (B-ALL).^{62,87} All VPs share the same model structure, and all have similar levels of malignant cells at the start of the trial. All VPs have parameter values that are within reported ranges, but the VPs differ in the values chosen within those ranges for some sensitive parameters, such as the malignant cell doubling rate. Some combinations of parameters were found to lead to treatment nonresponse (e.g., VP1), some to response (e.g., VP2), and some to relapse after initial response (e.g., VP3).

evidence. If a qualitative uncertainty persists (i.e., more than one hypothesis is consistent with relevant knowledge and data), its effect on the research question can be explicitly evaluated by maintaining multiple possible model structures and analyzing differences in predicted responses to various *in silico* experiments. This can be achieved through the use of virtual patients (VPs)^{21,33,36,41,42,60,68,80,81} – alternative versions of the model in which sensitive pathways or parameters are deliberately varied to explore the systemic effects of these differences.

In the interest of efficiency and focus, every effort should be made to assess the likely impact of a qualitative uncertainty as early in the modeling process as possible, using sensitivity analysis as well as scientific judgment. Proximity to the research target is one useful heuristic during model development. For example, qualitative uncertainty about the molecular target of a drug may merit close examination given likely implications on efficacy predictions, dosing decisions, or expected safety profile. Once the model is completed under one set of assumptions about qualitative uncertainties, sensitivity analysis can help identify sensitive mechanisms to focus on. If a parameter is identified as sensitive via sensitivity analysis, the mechanism(s) that the parameter is involved in should be assessed in more detail to see whether, for example, choosing a different equation form that is still consistent with data and knowledge would lead to a different outcome.

For example, to construct VPs representing two different qualitative hypotheses, one in which two signals have an additive effect and one in which two signals have a less than additive effect, one could write two versions of the effect equation and switch between them using a switching parameter that is off for one VP and on for another VP. Each of the VPs would then be calibrated to have appropri-

ate behaviors that meet all the qualitative and quantitative tests (see sections below). This process typically clarifies trade-offs between parameters or pathways and can lead to new insights into disease and therapy mechanisms. **Figure 3** illustrates the VP concept—mechanistic differences are encoded as combinations of different parameter values in different versions of the model (i.e., different VPs). The effects of these differences on outcomes become clear when a protocol of interest is simulated.

Often, the VP exploration can be used to identify experiments that would resolve an uncertainty. If multiple structural hypotheses are similarly consistent with data and knowledge, then the multiple VPs can be used to investigate the impact of the uncertainty on the research questions of interest, as further described below. For example, Tess *et al.*⁴⁰ used multiple VPs to assess the impact on glucose area under the curve of alternate hypotheses about the combined effect of a GPR119 agonist compound and endogenous GLP-1.

Recommended qualification approach

Relevant qualitative uncertainties must be explicitly documented, and resolution should fall into one of the categories listed in **Table 2**.

RELEVANT QUANTITATIVE UNCERTAINTIES ARE ASSESSED

Quantitative uncertainties are extremely common in mechanistic physiological models. The vast majority of biological systems are incompletely characterized in terms of rates, kinetic profiles, dose responses, absolute amounts, and other descriptive quantities. (Conceptually, we distinguish between uncertainty, in which the distribution of a parameter is completely unknown, and variability, in which the value of a parameter is known to have a given distribution. The latter is covered in the next section.) One goal of mechanistic modeling is to identify the subset of uncertainties that are sensitive (i.e., have an effect on outcomes)

Table 2 Possible qualitative uncertainty resolutions

Qualification aspect	Example questions that model qualification must address
Document and proceed with agreed-upon most likely hypothesis.	Appropriate if the outcomes are unlikely to change regardless of which hypothesis is used (e.g., if impact is local, distal to the focus of the research, and/or transient).
Simplify model structure to avoid modeling uncertain area explicitly.	Appropriate if possible without compromising the model's ability to address research questions.
Resolve definitively (i.e., eliminate all but one hypothesis through data analysis and/or modeling).	Appropriate if model predictions relevant to the research question are sensitive or likely to be sensitive to the uncertainty.
Maintain multiple hypotheses (VPs) in model to explore the systemic impact on biomarkers or endpoints of different hypotheses explicitly.	Appropriate if model predictions relevant to the research question are sensitive to the uncertainty and if more than one hypothesis satisfies all constraints.

VPs, virtual patients.

and material (i.e., impact drug development decisions). Prioritization of sensitive and material uncertainties can inform experimental approaches to help resolve the uncertainty and reduce risk in drug development.

Typically, an initial model is constructed using a set of baseline parameter values that bring the model in compliance with qualitative and quantitative data and knowledge constraints (see sections below). Uncertainties are tracked and systematic sensitivity analysis (see ref. 78 for examples of techniques) is used to identify the subset of processes and associated parameters that have a significant impact on outcomes. For example, Maxwell and Mackay⁴⁶ used sensitivity analysis in a model of skin sensitization to identify the pathways that most contributed to antigen-specific T-cell expansion (the biological biomarker of greatest interest) vs. to the local lymph node assay sensitization index (the gold standard biomarker) and found only partial overlap in the set of key pathways, suggesting that the gold standard biomarker was not as biologically relevant as previously thought. In another application, Benson *et al.*⁸² used sensitivity analysis to identify sensitive druggable targets in a systems pharmacology model of the nerve-growth factor pathway.

After sensitivity analysis, VPs can be constructed in which sensitive parameters take on different values. Each VP is a targeted, complete alternative hypothesis of the biological system, meeting all relevant qualitative and quantitative constraints, such as responding appropriately to existing treatment protocols and diagnostic tests. Conceptually, VP development therefore often involves making changes to sensitive parameters and making compensatory changes elsewhere in the system (e.g., in a redundant pathway) to bring the VP in compliance with the constraints. Note that VPs are not created by simply drawing values from the possible parameter space, because a randomly drawn collection of parameters would generally fail to satisfy the constraints that a valid VP must satisfy. (If sampling techniques are used in the VP creation process, the candidate VP must be tested and refined or discarded if it fails to satisfy the testing criteria.) The set of VPs should each satisfy all constraints while collectively being as different from each other mechanistically as possible.

Once created, VPs can then be used to investigate questions of interest, such as:

- How do different hypotheses about disease pathophysiology (represented by different VPs) affect the choice of optimal dose or protocol?
- What biological features distinguish responders from nonresponders?
- What biomarkers could be used to identify these patients clinically?

The optimal or necessary number of VPs depends on the research context and the degree of relevant uncertainty and variability inherent in the biological system. At a minimum, enough VPs should be created to explore the range of possible values for any parameter sensitive enough to have a material impact on the outcome of interest. It is often instructive to create VPs close to the extremes of

possible parameter values to assess whether uncertainty in the parameter is likely to pose a risk. If VPs with parameter values at opposite ends of the feasible range have similar responses to the drug of interest, then resolution of the uncertainty is probably not necessary. In such a case, it may also not be necessary to create a larger set of VPs with intermediate values in the parameter under investigation.

Generally, modeling projects investigating questions in discovery or early development, such as go/no-go on a new target should focus VP creation on highly sensitive qualitative and quantitative uncertainties, but do not require the collection of VPs to match the distribution of observed clinical outcomes. Modeling projects investigating questions in later development or lifecycle management typically require more VPs covering the range of clinical outcomes and may necessitate creation of a virtual population whose distribution matches clinical data.

Recommended qualification approach

Identify sensitive quantitative uncertainties (e.g., through sensitivity analysis). The impact of sensitive uncertainties on research questions or drug development decisions can be investigated through the use of alternative VPs.

MODEL CAPTURES RELEVANT KNOWN PATHWAY VARIABILITIES

Managing variability is one of the great challenges in drug development. In this tutorial, we consider two kinds of variability: pathway and outcome variability. Pathway variability is known interpatient variability that occurs at the mechanistic pathway level, such as when different etiologies give rise to a similar disease state. For example, the extent to which patients' asthma is driven by eosinophils vs. neutrophils, known to vary across patients, would be considered a pathway variability. Outcome variability refers to the variability in observed outcome variables.

Mechanistic physiological modeling seeks to identify and elucidate causal mechanistic links. Known pathway variability is of interest because it is one of the factors giving rise to outcome variability. In the asthma example, it is now known that the eosinophils vs. neutrophils pathway variability does account for some of the outcome variability in patients' responses to particular drugs, because the role that the drug targets play in patients' pathophysiology varies. Experts in any field have mental models of how pathway variability leads to outcome variability; a well-qualified physiological model makes these links explicit and testable.

As is the case for uncertainty, VPs are helpful in exploring the impact of pathway variability on outcomes. Indeed, a common use of mechanistic physiological models is the construction of alternative VPs with hypothesized pathway variabilities that could explain observed clinical outcome variability.

Recommended qualification approach

Identify sensitive known pathway variabilities through a combination of scientific research and model-based

sensitivity analysis. Investigate the impact of pathway variabilities on research questions of interest through the use of alternative VPs.

RELEVANT OUTCOME VARIABILITY IS REPRODUCED

Analysis of outcome variability in the mechanistic physiological modeling context strives to illuminate the biological sources of outcome variability once drug-specific variability has been accounted for. (Existing PK models can often be directly incorporated, and the impact of varying PK parameters in accordance with their established distributions can be assessed.) Typically, known PK and pathway variability account for some but not all of the outcome variability. Note that outcomes here include both clinical outcomes and subsystem outcomes, such as response of an organ or cell to a given protocol.

Clinical outcome variability is at the heart of many challenging questions in drug development, such as:

- Will a novel drug work well enough in a range of different types of patients?
- Is the variability seen in response to a competitor drug because of underlying pathway variability in patient pathophysiology or because of drug-specific properties?
- Could clinical biomarkers stratify patients by relevant pathophysiology and explain or reduce clinical variability?
- To what extent could clinical variability be explained by poor adherence to the protocol?
- Is combination therapy beneficial in all types of patients?

Mechanistic physiological modeling can help address these questions by improving understanding of the connection between mechanisms and outcomes. Investigations into the mechanistic sources of clinical outcome variability may be of interest even at early stages of drug development. For example, if there is a subset of patients who fail to respond to the current standard of care drug, understanding whether a novel drug may help these patients could be of great strategic value. In this scenario, it is the clinical variability in response to a different drug that the model should first focus on exploring. It should be noted, however, that the degree of emphasis on clinical outcome variability varies depending on the specific research context and data availability. See the following sections for approaches to reproducing outcome variability at the level appropriate for the research context. Lack of relevant clinical outcome data does not invalidate the model, it just implies a greater qualification emphasis on subsystem outcomes and on exploration of uncertainty and variability that could lead to clinical outcome variability and thus pose a program risk.

When outcome data are available for the drug under investigation or for mechanistically related drugs in the clinical population of interest, the model must demonstrate the ability to reproduce observed outcome variability. This is achieved by creating a number of VPs with diverse parameter values reflecting PK and pharmacological variability, known pathway variability, and uncertainty in sensitive pathways, within the ranges of observed data. Alternatively, the

model can be used to anticipate possible outcome variability by creating VPs that are as different from each other as possible while still observing relevant constraints. In either scenario, the model helps elucidate the connections between mechanisms and outcomes. In one application, Singh *et al.*⁶² used a mechanistic physiological model of acute lymphoblastic leukemia and created three VPs representing a prototypical nonresponder, responder, and relapser to conduct initial research and clarify mechanisms and sensitivities. The set of VPs was later expanded to cover the observed range of clinical variability and explore alternative dosing protocols in the intended patient population.⁶⁴

Recommended qualification approach

Demonstrate the ability of the mechanistic physiological model to reproduce the observed range of outcomes for protocols and outcomes that are relevant to the research context. Use a number of VPs appropriate for the research context.

MODEL RESULTS ARE QUALITATIVELY CONSISTENT WITH RELEVANT DATA AND KNOWLEDGE

Mechanistic physiological models give users the ability to simulate many different protocols and observe all variables at every timepoint. This presents extraordinary opportunities for qualitative testing, most commonly, visual checks of model results under many conditions and comparison to related data and expert knowledge (visual checks are also commonly utilized in statistical modeling^{83,84}).

Importantly, qualitative testing does not require specific types of data or sampling frequency. Many valuable and mechanistically relevant data are from only somewhat related populations (e.g., patients with rheumatoid arthritis in Asia vs. irritable bowel disease in America), animal data, etc. Although such data may not be suitable for strict statistical testing, they nonetheless contribute to the overall scientific understanding of disease mechanisms. It is appropriate to expect simulated model responses to be qualitatively consistent (e.g., similar time profiles, shapes of curves, relative responses to tests) with the data that are available and other knowledge, such as known differences between patients or species. Criteria that can be used to check qualitative consistency depend on the biology and the data available and may include, for example, expecting to see a similar time profile of response in patients with rheumatoid arthritis and patients with irritable bowel disease, or expecting similar relative responses to different therapies in patients with moderate vs. severe disease.

Qualitative testing includes comparing VP responses to reported data ranges for a variety of protocols, and it also provides additional constraints when data were sparsely sampled. For example, it may be quite reasonable to expect each VP's time trajectory of response between the measured points to be smooth (i.e., VPs with significant transient excursions or oscillations could be considered invalid, despite not violating any explicit data constraints).

Typically, experts have many more test criteria in mind than they can easily enumerate and articulate. An

interactive process of running simulations, showing graphical results, and asking for feedback from experts is extraordinarily useful in eliciting a more complete set of conditions that the model should meet in order to be considered valid. Qualitative testing thus ensures that the model is consistent with the known science and the experts' state of knowledge, not just with specific data points.

Recommended qualification approach

Identify and document qualitative test criteria with expert input and feedback. The most common qualification technique is visual check of time charts.

MODEL MATCHES RELEVANT PRESPECIFIED QUANTITATIVE TEST DATA

Perhaps the most obvious measure of model quality is its ability to reproduce relevant test data. Quantitative testing can be done at different levels, depending on the research context and data availability.

During model construction, a vast amount of data is identified and used to inform choices of parameter values as well as to serve as quantitative test data at the subsystem or whole-system level. For mechanistic physiological models that are deterministic in nature and built for the purpose of elucidating mechanisms, not optimizing parameter estimates, separating build and test datasets is not generally appropriate or useful. The most efficient workflow is one where all available data are used to first build the mechanistic pathways and then test that the model simulations reproduce the system-level data (e.g., clinical outcomes).

It is gratifying when a mechanistic model predicts results that are subsequently shown to be in agreement with new data. This is an indication that the representation of biology implemented in the model can produce relevant behaviors and extrapolate beyond the clinical tests that were already conducted. However, one should be cautious in interpreting such an outcome as "validation." These models are typically much too complex to be confidently validated with any single dataset. Further, unless the new data are for an experiment testing a mechanism for which no data were available before, this is a weak validation of a deterministic model, because the mechanism would already have been tested with existing data. Finally, if the new data are for a new mechanism, then should the model fail to match the new data, this would be an indication that some aspect of biology was not previously well understood and thus an opportunity to learn and improve the model. Thus, although matching new data certainly adds confidence in the model, this type of "validation" is neither strictly necessary nor sufficient, and should therefore not be overemphasized in the overall model qualification plan.

Relevant quantitative testing for mechanistic models often involves ensuring that every VP's simulated outcomes fall within a prespecified range of reported outcomes (e.g., within two SDs of the mean). This is done using many different types of test protocols that perturb different parts of the system, thus ensuring that each VP's set of model parameters can reproduce appropriate outcomes in response to a range of relevant protocols. For example, every VP in

the model developed by Tess *et al.*⁴⁰ was tested against sitagliptin, exenatide, metformin, and glyburide, as well as meal tests and infusion protocols. If a research goal is investigation of the mechanistic drivers of outcome variability, it may be of great interest to create a set of VPs that collectively span the range of observed outcomes reported, especially in clinical trials of drugs with mechanisms of action that are mechanistically related to the compound under investigation.

When the research context requires it, simulation-based quantitative statistical testing can be performed (e.g., to support clinical trial simulations using populations of VPs to reproduce outcome distributions). Clinical data for quantitative testing must meet the following relevance criteria:

- The clinical data population is similar to the VP population.
- The protocol is relevant to the research context.

Randomly sampled VPs with relevant PK and mechanistic variability can then be simulated using the appropriate protocol, and simulation results can be used for a visual predictive check.⁸³

Note that statistical testing is never sufficient for assessing the fitness-for-purpose of a mechanistic physiological model, nor does lack of data appropriate for statistical testing mean that the model cannot be qualified. Confidence in the model primarily derives from the fact that it draws on many sources of data and knowledge to construct a coherent mechanistic representation of the biological system under investigation²⁸ and that it addresses all of the other criteria of the MQM.

Recommended qualification approach

Identify and document the relevant data and criteria. Quantitative testing criteria depend on the research context and the data available.

DISCUSSION

Drug compounds interact with complex biological systems that include redundancies, nonlinearities, and feedback loops. Data and understanding of the biology are always limited. Patient variability further complicates the picture. Pharmaceutical development challenges multiple researchers in different disciplines to integrate vast amounts of different types of data and prior knowledge into a coherent framework to arrive at an optimal course of action for advancing therapeutics that will ultimately make a meaningful difference in patients' lives.

Mechanistic physiological modeling is a scientific approach that utilizes understanding about biology to facilitate interpretation of data and exploration of hypotheses in a model that reproduces relevant complex behaviors. The MQM presented here is a customizable, complete, and practical approach for determining if a mechanistic physiological model is qualified to support drug discovery and development (i.e., if it is "fit for purpose"). The current discussion has been mostly limited to the use of the MQM for qualifying mechanistic physiological models for drug development, the context in which this

method has proven successful. Recent publications suggest that the concepts are portable to QSP more broadly or other modeling methodologies.^{4,17,70,85} Similarly, the MQM framework can also be applied to model qualification in other contexts, such as academic or regulatory. It should be noted that the same model in different contexts may require additional or different qualification if the research question or intended model use is different. For example, a regulatory decision may require more extensive model qualification and documentation than an internal decision. The MQM framework emphasizes the use of relevant criteria, and relevance depends on the research context.

Pharmaceutical development organizations increasingly recognize the need to integrate across functional areas and make better use of data and knowledge that have for too long been siloed in different parts of the organization. Mechanistic physiological models can be ideal tools for supporting these strategic initiatives provided that team members have confidence that the models are fit for purpose. The MQM laid out here can serve as a planning, communication, and documentation framework. In the planning and scoping stages, the MQM clarifies the process of model development and qualification and ensures alignment on the research context. For a model to be qualified as fit for purpose, it is critical that that purpose be explicitly agreed upon, documented, and periodically revisited. Clarity of the research context then facilitates the process of making scope decisions and getting agreement on the testing criteria and overall qualification plan. During model development, relevant biological discussions, decisions, assumptions, uncertainty, variability, and tests against data and knowledge should all be presented to the team in the context of the MQM framework and documented. In this manner, the information that the team needs to assess model quality is generated along with the model. Consistent and careful application of the MQM can thus ensure that model-based research is seen as relevant and actionable for drug development.

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