



Moving from Animal Models to the Clinic

A Complimentary Market Research Study
as included in the Animal Models for Therapeutic Strategies Report

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Modeling and Simulation

Computer Modeling and Simulation is Complementary to, but Cannot Replace, Animal Studies

In silico modeling and simulation is used throughout the drug development process, from discovery through late-stage clinical trials. Computer simulation can reduce the numbers of animals needed in the research and preclinical stages of drug discovery. An example is the use of *in silico* ADME modeling. *In silico* modeling and simulation also helps translate preclinical data into the design of human clinical trials, as discussed in the next section.

Various groups have proposed replacing, in whole or in part, research and preclinical studies using animal models (especially mammalian models) with a combination of *in vitro* assays, computer simulations, and noninvasive or minimally invasive human studies. Such human studies might involve imaging or early stage human clinical trials that use microdosing. Microdose studies use doses of a drug that are too low to induce pharmacologic effects and are designed to evaluate pharmacokinetics (PK), pharmacodynamics (PD), and/or mechanism of action (MOA) of a drug, or to evaluate imaging of specific targets. The risks to human subjects in such studies are deemed to be minimal. Microdosing or Phase 0 clinical trials must be approved by the FDA under an Exploratory IND in the United States, or by other regulatory agencies in other jurisdictions.

One group with an interest in replacing animal studies is, of course, animal rights activists. However, others, believing that “animal models are not very predictive anyway” propose finding ways to replace them with computer modeling, *in vitro* studies, and minimally invasive human studies.

However, a computer simulation is only as good as the data that is used to program it. Creating a simulation of a physiological or pathophysiological process requires knowledge of the physiological or pathophysiological process itself. Data on such processes must come from biological studies, which in the vast majority of cases are animal studies. *In vitro* biochemical and cellular assays can also help reduce the numbers of animals used in the early stages of drug discovery, as well as help researchers design better animal studies. However, as discussed **CHI’s Insight Pharma Report, Animal Models for Therapeutic Strategies**, even such sophisticated and advanced systems as iPS cells and differentiated cells and disease models derived from iPS cells cannot replace animal models, but are complementary to them.

As we discussed in CHI’s report, a major reason for why animal models are poorly predictive is that we understand relatively little about normal and disease biology. Also, as shown by recent research in human genetics and genomics of disease, we understand much less than we thought we did. There are levels of cellular and organismic regulation that have only been discovered within the last few years, and non-coding DNA (which is mainly of unknown function) makes major contributions to the genetics and biology of disease. Computer simulations are, of course, completely unable to deal with these unknown factors, since no one can program them into a computer model. As shown by the seemingly continual discovery in recent years of previously unknown classes of small regulatory RNAs (some of which have been discussed in previous chapters of this report), and the unknown functions of most evolutionarily conserved non-coding DNA sequences, there are likely to be further layers of regulation that are yet to be discovered.

However, since humans share the vast majority of the recently discovered layers of regulation and non-coding DNA sequences with other mammals, researchers can use mammalian models to learn about these new factors in disease biology. They can also use the new knowledge gained from these animal studies to design, in an iterative manner, more predictive animal models. They can also design “humanized mouse models” that may be more predictive of clinical results in certain diseases than more conventional mouse models.

As discussed in the report, researchers can move toward development of mammalian models in species other than the mouse that may be better models of certain diseases than are mouse models. Researchers can also maximize the predictive value of animal models with respect to efficacy by testing a drug candidate in more than one animal model.

Even with animal models whose predictive value is less than what is desired by researchers, animal testing serves to lower the risk of bringing drug candidates into clinical trials. This is especially true with respect to safety, but also with respect to efficacy. Lowering the risk of administering an unsafe drug or unsafe levels of a drug to humans is the main reason for testing drugs in multiple mammalian species, as discussed in Chapter 1. The initial dosage of a drug in first-in-humans clinical trials is based on data from these preclinical studies, such as the NOEL (No Observed Effects Level) or the No Observed Adverse Effects Level (NOAEL). NOEL is the highest dose that gives no biological effects of any kind, and NOAEL is the highest dose that gives no adverse effects.

An additional issue with computer simulations is that the processing power of computers available to researchers, even in Big Pharma companies, may be too limited to model many physiological and disease processes such as those involved in neurobiology. For example, modeling of the neurobiology of the brain may require use of the most advanced supercomputers. Pharmaceutical/biotechnology researchers usually do not have access to these machines nor understand the computer science and mathematics necessary to design and interpret complex simulations that require supercomputing. Even with access to supercomputers, the data programmed into them are based on animal studies.

As for minimally invasive human studies via imaging or microdosing, any human testing of a drug of course requires administering that drug to humans. Animal testing (*e.g.*, determining the NOAEL) is necessary to reduce the risk of administering unsafe drugs or unsafe levels of a drug to humans, and regulatory agencies require such animal testing even for microdosing studies. As we discussed in a previous Insight Pharma Report,¹⁰ new approaches to early stage clinical trials (*e.g.*, proof-of-concept trials and Phase 0 studies that employ microdosing) can be used to reduce the risk of drug attrition in Phase II and III, including risks due to deficiencies in the “target validation paradigm” of drug discovery and due to poorly predictive animal models. However, animal model studies are necessary to design these trials and are also a crucial component in reducing late-stage attrition. Another means of reducing the efficacy risk of animal model studies is the use of translational biomarkers, as discussed in CHI’s Insight Pharma Report, *Animal Models for Therapeutic Strategies*.

Computer Modeling and Simulation for Moving From Animal Models to the Clinic

Allometric Scaling: Determining the Human Equivalent Dose (HED)

An important issue in moving from mammalian models in preclinical studies to human clinical trials is determining the human equivalent dose (HED), based on extrapolation of the safe starting dose in an animal. This process is known as “allometric scaling.” The FDA recommends that when selecting a starting dose for first-in-man studies, the NOAEL be used to determine the human equivalent dose (HED), via normalization to body surface area (BSA), in milligrams of drug per meter squared (mg/m²) of body area. However, it is difficult to measure body surface areas. Therefore, in its 2005 Guidance for Industry on estimating the maximum safe starting dose in initial clinical trials,¹⁴⁹ the FDA gives conversion values for determining a HED in milligrams of drug/kilograms of body weight (mg/kg) from an animal dose in mg/kg, based on scaling via mg/m². For example, one would divide a mouse dose (*e.g.*, the NOEL) by 12.3 or a rat dose by 6.2 to get a HED for an adult human. The use of allometric scaling based on body surface area

stems from decades of research, which has shown that such biologically significant factors as blood volume, amount of plasma proteins, oxygen utilization, and renal function are proportional to body surface area in various mammalian species.

When determining the maximum recommended starting dose (MRSD) for first-in-human clinical trials of new molecular entities in adult healthy volunteers, the FDA Guidance recommends that researchers first choose the most appropriate mammalian species for making this determination. The default position is that the most appropriate species is the most sensitive species in terms of NOAEL. However, for some drugs, the most appropriate species may be chosen on the basis of physiological or other biological similarities to humans that are specific to the drug (*e.g.*, the presence of relevant receptors, ADME parameters, the nature of the dose-limiting toxicity).¹⁴⁹ The agency recommends that researchers first use the NOAEL of the most appropriate species and determine the HED by multiplying it by the appropriate allometric scaling factor as discussed previously. Researchers should then apply a safety factor, which allows for: variability due to uncertainties in whether humans might be more sensitive to a drug than animal models, difficulties in determining some toxicities (*e.g.*, headache, mental disturbances) in animals, differences in target amounts or affinities, unexpected toxicities, and differences in ADME between the animal model and humans. The default safety factor is 10.

Allometric scaling for determining starting doses is not a perfect method. Dosing based on body surface area does not take into account the process of drug elimination or differences between human subjects with respect to physiological, biochemical (*e.g.*, expression of drug metabolizing enzymes), and other factors (*e.g.*, age and gender).¹⁵⁰ These factors may become more important as clinical trials move from drug testing in healthy volunteers (in Phase I) to drug testing in patients (in Phase II). (For cancer drugs, for example, patients are also used in Phase I trials.) Researchers would therefore need PD/PK models for determining dosages, rather than simple allometric scaling. Moreover, the overriding concern of the FDA in publishing its guidance document for determining first-in-man doses is safety. However, in Phase I clinical trials, drug developers not only want to assess safety, but also PK and PD, and for some diseases (especially cancer), in which patients rather than healthy volunteers are subjects in Phase I trials, developers (and patients) want to get a first look at potential efficacy in humans as well. Therefore, drug developers usually wish to use PK/PD modeling in the design of Phase I trials rather than simply rely on NOAEL determination and allometric scaling. However, NOAEL data may be incorporated into a PK/PD model to determine the probability of achieving efficacious exposure levels of a drug that are well below exposure levels that produce toxicity. Allometric scaling may also be incorporated into PK/PD models to help predict the effects of the drug in humans on the basis of its effects in animal models.

Pharmacokinetic/Pharmacodynamic (PK/PD) Modeling

Traditionally, using animal model data to determine the starting dose or dose range for use in first-in-human clinical trials was mainly empirical. However, with the industry's experience of failure of many late-stage clinical trials due to suboptimal dosing, and with the growing cost of drug development, companies have moved to the use of pharmacokinetic/pharmacodynamic (PK/PD) modeling. This is aimed at predicting how a drug will perform in patients with respect to dose/concentration relationships and how these may affect safety and efficacy.

Pharmacokinetics (PK) involves determination of the absorption, distribution, metabolism, and elimination properties of a drug. Pharmacodynamics (PD) refers to the study of the biological effects of a drug in the body, including the relationship between the drug's concentration and its effects. PD intrinsically involves the mechanisms of action of the drug, such as receptor or target binding, post-receptor effects (*e.g.*, signal transduction), and interaction of the drug with other molecules in the body. The combination of PK and PD helps explain the relationship between a drug's dose and response in patients. PK/PD modeling and simulation (M&S) is used at every stage of drug development, in order to enable decision making by drug developers and manage clinical risk. For example, it may be used in the design of Phase II clinical trials based on data

from Phase I trials, or in the design of Phase III trials based on Phase II data. In this section, we focus on the use of PK/PD M&S in designing Phase I trials based on preclinical animal model data.

In the research stage, drug developers may use animal studies to determine the mechanism of action of a drug [e.g., What is the drug's target? What are its biological effects once it binds to the target (e.g., Agonist or antagonist? Intracellular signaling, inducing apoptosis in tumor cells, inducing insulin secretion in pancreatic beta cells, inhibiting unwanted immunological reactions, etc.)? Are there off-target effects and/or on-target adverse effects, etc.?] They may also use animal studies in lead optimization. In preclinical animal studies, researchers may obtain data in several different animal models relating to the drug's efficacy and potency, its clearance, its bioavailability, and other PK/PD data. They also use animal studies to determine the drug's safety and toxicity characteristics (e.g., the NOEL). As discussed in CHI's report, ADME and toxicity screening may also be done in cellular models and/or in such non-mammalian systems as zebrafish larvae. However, pivotal preclinical data for regulatory agencies must be obtained in mammalian systems. Data from cellular and zebrafish larva models are used for such purposes as screening out compounds that are toxic or have poor ADME properties early in development. However, PD/PK M&S may include data from *in vitro* studies, such as binding affinities of drugs to target molecules. Such data are incorporated into PD/PK models along with quantitative PD and PK data from mammalian models, in order to predict human PD/PK parameters and determine doses that are likely to be safe and efficacious in humans.

In developing PD/PK models for moving from preclinical studies to first-in-man trials in testing a NCE (new chemical entity), researchers need to assess the drug's efficacy and safety characteristics in animal models. These may include actual efficacy and safety outcomes, as well as biomarker responses that may be predictive of efficacy or safety. Researchers also need to determine the dose range that should be studied in early clinical trials.

Sometimes researchers have a great deal of information on the PK, PD, efficacy, and safety of approved (comparator) drugs in the same therapeutic class as the new drug to be clinically tested. In these cases, they take an empirical approach to building a PK/PD model based on the behavior of the comparator drugs and the new drug in animal models, as well as on the behavior of the comparator drugs in humans.¹⁴⁸ Allometric scaling is used to convert the calculated efficacy and safety parameters of the drug in animals to expected values in humans.

If there are no comparator drugs that may be used to construct empirical models, then researchers perform mechanistic modeling PD/PK. Such models are designed to characterize processes that relate drug exposure to drug response. The models include terms that represent the distribution of target molecules in the body, the binding and activation of target molecules by the drug, and the resulting signal transduction and biological effects, including the interaction of drug effects with disease processes.

Several companies provide software for developing PK/PD models in the pharmaceutical industry. A leader in this field is Pharsight (St. Louis, MO). Pharsight's WinNonlin is the industry standard for PK/PD analysis. In addition to its extensive library of built-in PK, PD, and PK/PD models, WinNonlin supports custom models developed by the users. In June 2009, Pharsight announced the launch of its next-generation version, Phoenix WinNonlin. GastroPlus, an ADME simulation program developed by Simulations Plus (Lancaster, CA), is also used by pharmaceutical researchers in preclinical and clinical PK/PD modeling.

Modeling and Simulation at Novartis

Several large pharmaceutical companies, including Pfizer, GlaxoSmithKline, Lilly, and Novartis, have been implementing PD/PK M&S. In particular, Novartis has established a dedicated M&S department, which works with the company's drug development therapeutic franchises. It is

headed by Donald Stanski, MD, who is Vice President and Global Head of Modeling & Simulation at Novartis. Dr. Stanski is the former Vice President of Scientific and Medical Affairs at Pharsight.

Novartis' M&S department has a very broad scope, ranging from modeling of signal transduction pathways and safety modeling (*e.g.*, cardiac safety), to clinical trials, economic modeling, and decision analysis. One aspect of the department's activity involves development of models to help move drug candidates from preclinical into first-in-man studies.

For example, Novartis researchers completed preclinical studies of a monoclonal antibody drug designed to treat spinal cord injuries. However, the human spinal column has a very different geometry and fluid dynamics than spinal columns of animal models. Therefore, traditional allometric scaling between animal models and humans is not likely to give useful results. The Novartis M&S team therefore used a biophysical modeling platform to model the human spinal cord and its surrounding tissues. The goal of this modeling effort was to describe the geometry of the spinal canal and the relevant transport properties that are applicable within that geometry, when the monoclonal antibody drug is injected into the intrathecal space at a certain point in the spinal canal. A key question is whether the drug will reach the site of action. Variables within the model included the type and orientation of the syringe, the location along the spinal canal, the rate of injection, and whether the patient was standing or lying down.¹⁵³ This example illustrates that moving from animal models to humans in drug development is not always a straightforward process. Various types of computer models may be useful in improving the way in which drug developers plan first in-human clinical trials based on preclinical data.

Entelos

Entelos (Foster City, CA) develops predictive *in silico* mechanistic models of human disease. These are meant to help pharmaceutical researchers reduce the time and cost of carrying out drug discovery and development operations, ranging from target validation through Phase III and IV trials, and improve the success rate of clinical trials. The company's business model involves entering into R&D partnerships, research agreements, licenses, and strategic alliances with pharmaceutical and biotechnology companies. Entelos focuses on building dynamic, large-scale computer models of human physiology and disease, based on its proprietary PhysioLab technology. Models integrate relevant genomic, proteomic, biochemical, physiological, and environmental data that pertain to a disease or therapeutic area, with the goal of predicting clinical responses to potential drugs or other therapeutics. Data for constructing these models come from the scientific literature, the expertise of Entelos' researchers and scientific advisors, and proprietary information from Entelos' clients.

These models do not include every signaling pathway in the body, which would require the use of too many sets of differential equations that would be beyond the ability of even advanced supercomputers to manage. Moreover, there are significant unknowns in biological systems that could not be included in any model. Therefore, many aspects of cellular physiology are modeled by aggregating the functional effects of underlying pathways and networks and representing them at a higher level of detail. For example, a model may represent a cell type that under specific conditions produces certain cytokines at specific rates. This does not model the complex intracellular pathways that are involved in producing the cytokines. However, if a client is interested in a certain target, the pathways in which that target participates may be modeled in detail. Even with this approach, gaps in knowledge remain. The models therefore explicitly represent these knowledge gaps and aim to enable researchers to define the gaps and develop experiments to resolve them.

Based on PhysioLab technology, Entelos has developed models called "virtual patients." These models of human disease represent the range of patients affected by a particular complex disease, including the effects of unknown genetic factors and behavioral/environmental factors that are involved in each disease. Included in these modeling efforts is the development of tests to validate the virtual patients as behaving like real human patients in terms of their physiological readouts. Disease-specific PhysioLab platforms include systems for diabetes and obesity and

immune/inflammatory diseases including asthma and rheumatoid arthritis. Within each of these disease platforms, Entelos can create a potentially unlimited number of virtual patients, each of which represents a subpopulation of actual human patients. A virtual patient may represent a known or hypothesized cause of disease. Using simulation experiments, researchers can test marketed and experimental therapies to predict a patient's likely response to treatment.

Virtual patient models are designed to represent human disease physiology, not animal models. However, one of their uses is to design early stage human clinical trials. Thus the models may be used to move from animal studies to the clinic, which is the focus of this chapter. Virtual patient models may also be used in drug discovery, for example in target validation or to predict which types of drugs might work best in various groups of patients prior to discovery-stage or preclinical animal studies. The results of the readouts of the virtual models would then be used to design animal studies. Entelos' virtual patient models are being widely applied across the pharmaceutical industry.

Among Entelos' partners are Pfizer, Merck, Novartis, AstraZeneca, Bayer, Bristol-Myers Squibb, Lilly, Johnson & Johnson, and Roche.

Entelos/American Diabetes Association virtual NOD mouse model

There is one case, however, in which Entelos' PhysioLab platform has been used to design a "virtual animal model." This is the design of a virtual non-obese diabetic (NOD) mouse by Entelos working in collaboration with the American Diabetes Association. The NOD mouse strain is a longstanding (first reported in 1980), standard model of type 1 diabetes, which is a multigenic disease that involves T cell-mediated autoimmune destruction of the insulin-producing pancreatic beta cells. Since there have been tens of thousands of published experiments done using the NOD mouse, the diabetes research community has voluminous data on this animal model. As with the human disease, causation of diabetes in the NOD mouse involves multiple genetic determinants. Researchers have been using this mouse strain to study the genetics of type 1 diabetes both in mice and humans.

Human type 1 diabetes is currently incurable, and patients (who typically develop the disease in childhood) must undergo lifelong injected insulin therapy. Patients may also develop serious diabetic complications, including cardiovascular or kidney disease or foot ulcers requiring amputation. Researchers in academia and in numerous companies have long been working on developing potential curative treatments, especially drugs to be administered early in the course of development of the disease, before the pancreatic islet cells are completely destroyed. In the research and preclinical stages, these drugs have usually been tested in NOD mice. Frustratingly, many drugs that showed efficacy in the NOD mouse failed in the clinic. An important motivation for development of the virtual NOD mouse is to attempt to improve the predictiveness of this animal model. (Entelos has also developed virtual patient models of human type 1 diabetes.)

Entelos' virtual NOD mouse is a large-scale, dynamic mathematical model of the female NOD mouse.¹⁵⁶ (Female NOD mice develop diabetes earlier and at a higher rate than male mice.) Construction of the virtual model was made possible by the large amounts of data available on this well-studied animal model. The virtual NOD mouse model represents components of the immune system and beta-cell physiology that are involved in the pathobiology of type 1 diabetes. Thus, as with other PhysioLab models, the virtual NOD mouse only models body systems that are known to be directly involved in the disease. Virtual NOD mice are constructed to represent variations in disease phenotype seen in actual NOD mice. These specifically include variation in disease progression rate, encompassing early, average, and late-onset disease. These individual virtual NOD mouse models thus are analogous to individual virtual patients that are designed to represent subsets of patients with complex diseases that have particular disease phenotypes.

Entelos researchers have used the virtual NOD mouse models to simulate treatment with various therapeutic regimens. For example, researchers simulated treatment of NOD mice with

exogenous interleukin-10 (IL-10).¹⁵⁶ When recombinant IL-10 was used to treat actual NOD mice, it was ineffective in preventing the onset of diabetes in mice with early disease onset. The Entelos researchers, using virtual NOD mice made to represent either early or average disease onset, reproduced the results seen with the actual animal model. The virtual study also indicated that the reason why IL-10 treatment of mice with early-onset disease was not efficacious was because autoimmune T cells had already infiltrated pancreatic lymph nodes and islets, and there was a high level of beta-cell destruction. According to the simulation, earlier treatment with IL-10 would protect the mice from developing diabetes. Entelos researchers have also been using the virtual NOD mouse to model other drug treatments, with the goal of helping researchers to develop new therapeutic regimens (drugs or drug combinations) by predicting their behavior in actual mice. As with the IL-10 study, studies with the virtual NOD mouse suggest that timing and dosing of drugs or drug combinations are important in successful treatment of the mouse, and presumptively of human patients as well. The researchers must still test the putative therapies in actual mice, however, and develop strategies (perhaps in part using simulation models) to translate their results into potential clinical candidates and into clinical studies to determine optimal administration and dosing regimens.

In August 2008, Entelos and the American Diabetes Association offered free access to the virtual NOD mouse platform to all the latter's health professional members via Entelos' Web-based Realab. At the same time, Entelos reported that when it designed the virtual NOD mouse models, its researchers discovered subtle differences between mice and humans that may be relevant to translating results of animal studies and designing clinical trials. The company said it intends to increase researchers' access to Entelos' virtual mice and virtual patients. This would enhance researchers' ability to test potential type 1 diabetes treatments in these platforms and eventually be able to use fewer animals but design more definitive (and hopefully more successful) clinical trials on the basis of the animal studies.

Entelos, in collaboration with its scientific advisors, is extending its virtual NOD mouse effort by constructing a virtual *Idd9* mouse. *Idd9* is one of the genetic loci of the NOD mouse that is involved in the pathogenesis of type 1 diabetes. However, a congenic mouse strain, NOD.B10 *Idd9*, whose genome includes the *Idd9* locus of the NOD mouse but not the other NOD diabetogenic loci (although nearly all mice of this strain develop insulinitis [inflammation of the pancreatic islets]), is resistant to developing diabetes. Since many of the genes (and/or the pathways in which they are involved) are already included in the virtual NOD mouse model, this helps simplify construction of a virtual *Idd9* mouse. Entelos and its scientific advisors hope to use the virtual *Idd9* mouse in comparison with the virtual NOD mouse to help identify experiments that can be done in live mice to explain the diabetes resistance of the *Idd9* mouse.

Translational Biomarkers

Another tool that is increasingly important in moving from animal models to clinical trials is the identification and use of translational biomarkers (also known as bridging biomarkers). We discussed biomarkers (translational biomarkers in particular) in an earlier Insight Pharma Report, *Approaches to Reducing Phase II Attrition*.¹⁰ That report focused on the use of translational medicine together with improved drug discovery strategies to improve the productivity of pharmaceutical R&D. Biomarkers are central to the science and practice of translational medicine.

Biomarkers are biochemical, molecular, physiological, or anatomical parameters that measure or predict a human subject's or an animal's state of normal physiology or disease, or their response to a drug. Translational biomarkers are biomarkers that can be measured in different species, especially in animal models used in research and preclinical studies and in humans. Such biomarkers, which may be identified and qualified in the research stage, may be carried forward

into preclinical studies and then used in human clinical studies. These biomarkers can thus be used to link drug responses in animal models and in human subjects (normal volunteers in Phase I studies and/or patients in Phase II–IV studies).

Translational biomarkers may be efficacy, safety, pharmacodynamic, or stratification biomarkers. (Pharmacodynamic biomarkers measure target occupancy by a drug, and stratification biomarkers are used to select patients for inclusion in clinical trials or for drug treatment.) Examples of translational biomarkers given in the earlier report include markers of inhibition of a signaling pathway by a drug (which were pharmacodynamic and/or efficacy biomarkers, since they indicated that the drug was reaching its target and inhibiting a pathway controlled by the target) and gene expression signatures of drug inhibition (which were stratification biomarkers, indicating classes of patients who might benefit from treatment with the drug). These biomarkers were identified and used in animal model studies, then carried forward into clinical studies. Also discussed in the earlier report was a set of urinary protein biomarkers of renal injury that were identified and qualified by the Predictive Safety Testing Consortium in 2008 (www.cpath.org/pstc.cfm). These markers can be used in preclinical rat studies to support clinical decision-making and have been qualified for this purpose by both the FDA and EMEA (European Medicines Agency). The use of these biomarkers in clinical trials may be considered on a case-by-case basis, in order to potentially qualify them for use in monitoring drug-induced renal toxicity in patients. If they are so qualified, they will be translational toxicity biomarkers.

From the point of view of researchers engaged in mechanistic PD/PK modeling, biomarkers may be defined as quantitative measures that characterize a process lying along the causal path between drug administration and drug effect.¹⁵⁸ Such biomarkers may characterize genetic and phenotypic factors that affect drug responses, concentrations of a drug and/or drug metabolites, occupancy of a molecular target, activation of the target, and measures of relevant physiological and pathophysiological parameters. Since in the context of PD/PK modeling such biomarkers may be characterized and used in both preclinical and clinical studies, they (once qualified) would be translational biomarkers.

Conclusions

Pharmaceutical and biotechnology researchers have been increasingly applying PD/PK modeling, especially mechanistic PD/PK modeling, to all stages of drug development. This especially includes moving from preclinical animal studies to human clinical trials. These models are the most widespread and important computer-based mathematical models used in drug development today. In the transition from animal models to human studies, an important focus is dealing with differences between animal models and humans, not only with respect to size (*e.g.*, allometric scaling) but also with respect to other characteristics such as differences in metabolism and the heterogeneity of the human population. Other types of mathematical modeling, such as Novartis' biophysical modeling of the spinal column, may also be employed to deal with specific differences between animal models and humans that may be relevant to clinical trials of particular types of drugs (in this case, a monoclonal antibody drug to treat spinal cord injury).

Entelos' virtual patient models are being widely applied across the pharmaceutical industry—as adjuncts to wet-lab drug discovery studies, animal studies, and clinical trials—in order to improve the time and cost factors in drug development and the success rate of clinical trials. One of their uses is in the design of early clinical trials. Thus, for companies that use these models, they (in addition to PD/PK models) are involved in moving from preclinical to clinical studies. However, they are human physiology models, not “virtual animal models.” Moreover, they do not model deep molecular, biochemical, and cellular mechanisms of disease biology or drug action. Thus, especially for development of novel therapeutic

strategies and innovative drugs, they are at best adjuncts to wet-lab biology and animal model studies.

There is one model developed by Entelos that can be described as a “virtual animal model.” That is the virtual NOD mouse. This mathematical model is made possible by the extensive studies that have been carried out for over 30 years with the living NOD mouse and the diabetes research community’s acceptance of its usefulness and relatively faithful modeling of human disease. For animal models that have proven to be poorly predictive of human disease (*e.g.*, most preclinical cancer and CNS disease models, which are the extreme examples), creating such a virtual model would not be possible. The same is true for novel animal models that lack the extensive characterization of the NOD mouse.

Even in the case of the virtual NOD mouse, the model is an adjunct to real animal studies, not a substitute. However, it may enable researchers to use fewer animals and conduct more effective animal studies and hopefully more effective and successful clinical trials. Nevertheless, the usefulness of the virtual NOD mouse in enabling researchers to discover innovative drugs that achieve proof of concept in clinical trials, let alone reach the market, remains to be confirmed.

The general limitations of computer models for creating useful “virtual animal models” that can replace real animal models, discussed at the beginning of this chapter, remain valid. This is especially true given the increasing numbers of unknowns in the normal and disease biology of humans and other animals (*e.g.*, novel levels of cellular regulation, the “missing heritability” of disease phenotypes) that have been (and continue to be) revealed in recent years. Animal models, for all their limitations, can help us learn about these unknowns and apply what we learn to pharmaceutical R&D. Computer models cannot do so.