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WHITE PAPER

Quantitative Systems Pharmacology for Neuroscience Drug Discovery and Development: Current Status, Opportunities, and Challenges

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The substantial progress made in the basic sciences of the brain has yet to be adequately translated to successful clinical therapeutics to treat central nervous system (CNS) diseases. Possible explanations include the lack of quantitative and validated biomarkers, the subjective nature of many clinical endpoints, and complex pharmacokinetic/pharmacodynamic relationships, but also the possibility that highly selective drugs in the CNS do not reflect the complex interactions of different brain circuits. Although computational systems pharmacology modeling designed to capture essential components of complex biological systems has been increasingly accepted in pharmaceutical research and development for oncology, inflammation, and metabolic disorders, the uptake in the CNS field has been very modest. In this article, a cross-disciplinary group with representatives from academia, pharma, regulatory, and funding agencies make the case that the identification and exploitation of CNS therapeutic targets for drug discovery and development can benefit greatly from a system and network approach that can span the gap between molecular pathways and the neuronal circuits that ultimately regulate brain activity and behavior. The National Institute of Neurological Disorders and Stroke (NINDS), in collaboration with the National Institute on Aging (NIA), National Institute of Mental Health (NIMH), National Institute on Drug Abuse (NIDA), and National Center for Advancing Translational Sciences (NCATS), convened a workshop to explore and evaluate the potential of a quantitative systems pharmacology (QSP) approach to CNS drug discovery and development. The objective of the workshop was to identify the challenges and opportunities of QSP as an approach to accelerate drug discovery and development in the field of CNS disorders. In particular, the workshop examined the potential for computational neuroscience to perform QSP-based interrogation of the mechanism of action for CNS diseases, along with a more accurate and comprehensive method for evaluating drug effects and optimizing the design of clinical trials. Following up on an earlier white paper on the use of QSP in general disease mechanism of action and drug discovery, this report focuses on new applications, opportunities, and the accompanying limitations of QSP as an approach to drug development in the CNS therapeutic area based on the discussions in the workshop with various stakeholders.

Central nervous system (CNS) diseases such as depression, Parkinson's disease, and Alzheimer's disease (AD) are complex and usually involve dysregulation in multiple biochemical pathways. It is likely that these disorders are not separate isolated conditions but, rather, a series of entities with shared clinical phenotypes. Although there are pharmacological interventions with proven effectiveness on symptoms, there are very few disease-modifying therapies for CNS disorders. Possible explanations include the lack of quantitative and validated biomarkers and the subjective nature of many clinical endpoints, but arguably most important is the fact that highly selective drugs do not reflect the complex interaction of different targets in brain networks. Therefore, it is reasonable to suggest that an approach that embraces disease complexity and the importance of network organization in the CNS could provide a promising alternative to current drug discovery approaches.

One such approach may be quantitative systems pharmacology (QSP), which merges systems biology and pharmacokinetics (PK)/pharmacodynamics (PD).[1] The term *systems pharmacology* was originally defined in the context of drug discovery as "…the body-system-wide, predominantly molecular, characterization of drug-perturbed state relative to the unperturbed state."[2] This definition was expanded to include translational research and drug development by the National Institutes of Health Quantitative Systems Pharmacology workshop group in 2011, which defined QSP as "an approach to translational medicine that combines computational and experimental methods to elucidate, validate and apply new pharmacological concepts

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to the development and use of small molecule and biologic drugs.... to determine mechanisms of action of new and existing drugs in preclinical and animal models and in patients."[3]

The development of CNS QSP will be influenced by opportunities for growth in the following four different dimensions: (i) pharmacology focusing on the "system" (see **Box 1**), rather than single targets to encompass multiple scales in space and time; (ii) the development of new *in vitro* and *in vivo* model systems suitable for controlled experimental interventions useful for validating QSP predictions; (iii) expansion of multi-omic data sets to understand both CNS physiology and pathology (see **Box 2**); and (iv) the development of quantitative, predictive multiscale computational models, network architectures, and analytical approaches that can explain the experimental observations, predict optimized experiments to test hypotheses, and most important, support drug discovery and development to translate these insights into useful therapeutic interventions.

To evaluate the potential of a QSP approach to accelerate drug development for CNS disease and to understand why this approach has not been more widely explored in the CNS area, a group of institutes at the National Institutes of Health (NIH; led by National Institute of Neurological Disorders and Stroke (NINDS) and including National Institute on Aging (NIA), National Institute of Mental Health (NIMH), National Institute on Drug Abuse (NIDA), and National Center for Advancing Translational Sciences (NCATS) sponsored a workshop titled, "Quantitative Systems Pharmacology and Drug Discovery: Filling the Gaps in Current Models of the R&D Process for Neurotherapeutics," which was held July 26–27, 2017, in Bethesda, MD (https://meetings.ninds.nih.gov/Home/General/16746).

The overarching goal of the workshop was to assess the impact of a QSP approach to CNS drug discovery and development on therapeutic target discovery, identification of therapeutics using physiologically relevant screening platforms, and development of therapeutics through the use of platforms that synthesize information about the PK, PD, and toxicological attributes of the drug in preclinical and clinical settings. Additional goals of the workshop were to discuss the roadblocks and gaps in the application of a QSP approach and to identify opportunities to overcome these challenges.

Participants in the workshop were charged with providing their recommendations about the potential of a QSP approach to address the current challenges in CNS disease drug development. This article will review ideas and suggestions to further develop QSP into a useful modeling platform for supporting CNS research and development (R&D) projects.

Box 1 Spatial and phenotypical scales and classes operational in systems pharmacology and possibly defining the "system."

- Individual biomolecular species
- Molecular classes (from proteins and lipids to nucleotides)
- Organelles (mitochondria, nucleus, Golgi apparatus, endoplasmic reticulum, etc.)
- Individual cells
- Cellular classes (neural, vascular, humoral, muscular, epithelial, secretory, microbial, etc.)
- Fluids (from blood to mucus)
- Tissues (from blood vessels to brain regions)
- Organs (liver and heart to gastrointestinal and kidney)
- Systems (from circulatory to reproductive)
- Individuals (different species)
- Populations
- Environment

Box 2 The hierarchy of experimental animal and human models in systems pharmacology

- *In vitro*
  - o Biochemical measurements
  - o Two-dimensional cell biology on plastic
  - o Microphysiological systems (individual and coupled organoids, tissue chips, and organs-on-chips)
    - ■ Primary cells and cell lines
    - ■ Induced pluripotent stem cells
- *In vivo*
  - o Physiological
    - ■ Noninvasive (excretome)
    - ■ Minimally invasive (tracer imaging, biofluids)
    - ■ Invasive (*in vivo* electrophysiology)
  - o Behavioral
- Pharmaceutical challenges in humans
- Task-mediated imaging studies

THE CHALLENGE OF CNS DRUG DEVELOPMENT

Drug discovery and development in the field of CNS disorders is extremely challenging, as evidenced by the recent failures of well-designed amyloid immunotherapy trials in AD.[4] Success rates, defined as final regulatory approval of a drug, are much lower for CNS drugs than for other diseases[5] and hover around 7–8%. In AD the situation is even worse, with more than 240 clinical development projects having failed since the approval of the last medication in 2004.[6] This particularly low success rate has been attributed to multiple factors, including the general complexity of CNS disease, lack of complete understanding of biology, overreliance on a "predominant hypothesis" (e.g., beta-amyloid), the gradual onset and slow development of some CNS diseases, the lack of animal models with good predictive validity,[7] issues with target accessibility in the brain, the subjective nature of many clinical end points, the difficulties in developing useful biomarkers, and general design challenges with clinical trials. For example, CNS therapeutics can require 15–19 years to advance from discovery to regulatory approval,[8] which is almost the same as the duration of current patent protection rights (20 years).

The drug discovery and development process has been traditionally a focused, phased, and sequential "derisking" approach, where decision-making criteria drive advancement

to the next phase. Prior to the 2000s, activities in each phase were somewhat isolated from each other, where "hit" identification and lead optimization were focused primarily on efficacy and selectivity. Investigations of clinical failures identified poor PK characteristics and safety barriers as major issues limiting the success during this period.[9] Many pharmaceutical companies mitigated these issues by requiring early characterization of PK properties, such as volume of distribution, time to reach maximum plasma concentration, elimination half-life, area under the curve, bioavailability, absorption, brain penetration, and so on as part of the decision-making process. Moreover, rapid high-throughput *in vitro* tests for absorption, distribution, metabolism, and elimination (ADME) properties became a standard part of the drug discovery and development process, dramatically reducing the failures of small-molecule therapeutics for PK reasons. As a result, the rate of clinical failures as a result of unsatisfactory ADME characteristics was reduced from 40% to approximately 10% by the early 2000s.[8,9] Tailored ADME strategies for CNS drug development are used by drug discovery organizations to address brain-specific PK issues: blood–brain barrier (BBB) and its active transporters (most notably, p-glycoprotein), and specific *in vitro* and computational tools were developed and validated.[10] However, these developments unmasked another major underlying contributor to clinical failures, which was lack of efficacy. A recent meta-analysis based on data from 2011–2015 estimated that 68% of so-called "confirmatory" phase III CNS fail because of lack of efficacy[11] for which a new 5-R (right target, right tissue, right safety, right patient, right commercial potential) framework has recently been proposed, although not specific for CNS disorders.[12]

In the case of chronic neurodegenerative diseases, often many comorbidities converge in the elderly brain. In addition, genetic studies suggest a large number of pathways involved in the pathology. As a consequence, it is unlikely that selective interventions focused on a single pathway will be able to provide a substantial clinical improvement. Notably, a recent task force in AD has identified combination therapy as the only way to get substantial progress in the treatment of this devastating disease.[13] As the number of possible targets and drugs is increasing, the only practical way to prioritize successful combination therapies is to use computational QSP in a large, multiscale, well-validated platform.

Therefore, new approaches are needed to turn the tide of failed CNS trials. An NIH White Paper written by participants in an National Institute of General Medical Sciences Workshop on the topic of QSP[3] proposed that merging systems biology and PK/PD approaches in a QSP framework could greatly facilitate drug discovery and development for complex diseases that have not been defined by a single molecular target. In this context, systems biology would identify the most likely network of genes, proteins, or neural pathways from large data sets with a variety of environmental, chemical, or genomic perturbations (**Box 3**). In a complementary fashion, different predictive mechanism-based QSP models (**Box 4**) would allow the vertical integration of molecular pathways to clinical readouts based on domain expertise, drug kinetics, and pharmacology based on an integrative, multidimensional approach. Injecting a quantitative spatial and temporal dimension should lead to the identification of more realistic drug discovery targets, along with more efficient prediction of PK/PD relationships and clinical outcomes. This merged discipline could provide a useful humanized translational tool from target validation to rational polypharmacy guidance in clinical practice as long as predictions against human clinical data sets were used to continuously test and improve the model.

Ironically, the basis for the field of computational neuroscience and quantitative biological systems modeling is founded in the Hodgkin and Huxley neuron model, which was developed in the early 1950s. Despite this head start, the development and application of QSP approaches in neuroscience has been lagging behind most other therapeutic areas that in part can be explained by the complexity of the pathophysiology of diseases of the CNS, which resulted in a widespread belief that the brain may be "too complex to model." However, the low success rates, high development

Box 3 Omics data sets and data sources for QSP models

Data sets

- Genome
- Transcriptome
- Proteome
- Lipodome
- Metabolome
- Secretome
- Epigenome
- Connectome
- Electro-encephalography, magneto-electro encephalography
- Imaginomics

Techniques

- Next-generation sequencing
- Next-generation sequencing of RNA (including single cell)
- Chromatin accessibility assays
- Mass spectrometry (proteomics, lipidomics, metabolomics, etc.)
- Cytometry with time-of-flight mass spectrometry
- Genetically encoded reporters
- Electrical stimulation (cortical, depth, transcranial magnetic)
- Multielectrode arrays
- Imaging

Box 4 Different computational models used in QSP

- Statistical (including Bayesian)
- Boolean
- Temporal (ordinary differential equations)
- Spatio-temporal (partial differential equations)
- Agent based
- Integrative
- Empirical curve fitting
- Machine learning

costs, and stagnation in the translation of preclinical science to novel medicines for neurodegenerative disorders has emphasized the urgent need for novel R&D approaches, and in recent years there has been a marked increase in the application of model-based approaches in neuroscience drug discovery and development. Indeed, a recent industry survey highlighted neuroscience as the therapeutic area with the greatest growth opportunity for QSP.[14]

Such a multiscale computational modeling approach integrating the impact of molecular processes on neuronal circuit activity could provide an alternative solution to developing successful CNS therapies.

This article will provide an overview of the current state of QSP approaches in CNS diseases, illustrated with case studies from AD, Parkinson's disease, and schizophrenia.

EXAMPLES OF SYSTEMS PHARMACOLOGY IN CNS DISORDERS

Brief historical overview of computational QSP in CNS disorders

The computational neuroscience community covers areas from abstract agent-based models of reward[15] to biophysically realistic models of electrical and chemical signaling in neurons and networks of neurons. Much of this academic computational effort is directed toward understanding fundamental neuronal processes such as memory, plasticity, encoding, and computation without consideration of either the pharmacology and pathology specific to CNS therapeutics or constraints to models that could be imposed by clinical data. Other computational studies have been focused on mechanism of action in disease, which is particularly useful for drug discovery, including modeling tau site modifications,[16] the aggregation dynamics of misfolded proteins,[17] the effect of neuro-inflammation and neuronal health,[18] and a spatio-temporal model of neuroinflammatory processes secondary to amyloid plaque formation.[19]

Applications in clinical neurology include the progression of amyloid load in AD,[20] PD biomarkers after treatment with beta-site amyloid precursor protein cleaving enzyme (BACE) inhibitors,[21] and a mechanistic QSP-based analysis on failed amyloid-modulating trials.[22] Computational modeling has been used to probe a large numbers of possible combination therapies for AD.[23]

Multiscale systems pharmacology in AD

A major barrier to successful AD drug development is the lack of AD models that actually mirror the human disease process with sufficient accuracy to predict drug response. For instance, although many models revolve around β-amyloid and tau, a large number of individuals with high levels of AD pathology are cognitively normal.[24–26] Therefore, it is important to create models—cellular, animal, or computational—that go beyond these two pathological hallmarks and are related to clinical outcome (e.g., cognition). Here, a complementary multiscale approach to assembling computational models of cognition that are amenable to QSP approaches is proposed based on connecting data acquired at multiple biophysical scales—the genome, molecules, cells, tissues, and the whole brain.

The motivation for a multiscale approach is that from a computational perspective, the continued failures of clinical trials based on existing models can be conceptualized as an inability to accurately "scale" the molecular actions of CNS drugs up to the level of clinical outcome. For every AD clinical candidate, there was strong evidence for the efficacy of each (now failed) molecule at the level of molecular activity and animal models. Ultimately the drug did not trigger a sequence of events that spanned multiple biophysical scales to improve cognition in humans—an emergent function of all those biophysical scales. By modeling multiple biophysical scales simultaneously in a multiscale approach using QSP, it may be possible to examine the effects of molecular perturbations on systems that are closer to cognition, thereby providing guidance on how to structure data generation, experiments, and simulations to develop more predictive models.

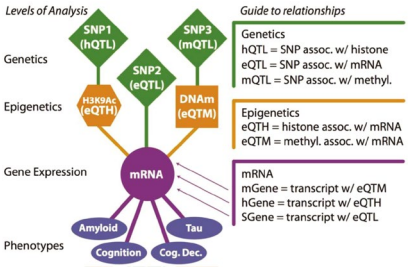
As an example, consider the clinical effects of BACE and gamma-secretase inhibitors in AD, suggesting a worsening of cognitive symptoms. By implementing detailed biological knowledge with a neurostimulatory effect of the short Abeta form with a negative effect of long Abeta forms, a mechanism-based QSP model first demonstrated that this differential effect was needed to explain three existing clinical data sets and further advanced a testable hypothesis on why high levels of inhibition of amyloid production would lead a worsening of cognition.[22]

To address the current inability to span spatio-temporal scales, one needs to acquire data at the genomic, molecular, cellular, tissue, and whole-brain level on the state of intermediate biophysical scales at various stages of AD. The collected data can then be coupled to each other sequentially (i.e., genetics to messenger RNA (mRNA) to protein to cells to brain network activity) but they can also be coupled by neural or humoral feedback loops that connect the functioning brain back to specific receptors, cells, brain regions, or other organs in the body. Some of these biophysical scales will be more directly supporting cognitive function, for instance, the number of synapses at the cellular scale, which is a close correlate of cognitive function in AD[25,27] compared to genetic variants, which are farther removed from direct cognitive function and brain activity but may act through multiple known or unknown intermediaries to affect synaptic function.

Building such multiscale QSP systems is therefore predicated on the availability of large multiscale data sets that measure cognition in addition to multiple molecular, cellular, and tissue-level phenotypes. Data generation efforts such as the NIH funded Accelerating Medicines Partnership–Alzheimer's Disease (AMP-AD)[28] are enabling multiscale AD modeling and other approaches by acquiring multiple types of omics data[29] from longitudinal cohorts such as the religuous order study and memory and aging project studies.[30]

As experimental data are typically acquired on a single biophysical scale, it remains challenging to link multiscale data types. However, several examples of practical multiscale experiments exist, often within the context of AD drug discovery (**Figure 1**). Each of the projects outlined combines different types of data that operate on distinct biophysical scales to identify behavior that is conditioned by multiple

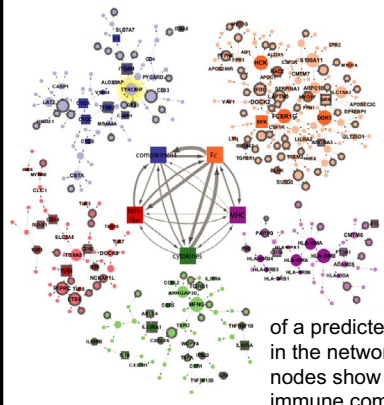
(a) Upstream gene prediction, via genetics, epigenetics & mRNA



Combining genetics and multiple omics into local regulatory networks leads to validated predictions of genes which drive cognitive phenotypes.

Projects span biophysical scales (below)

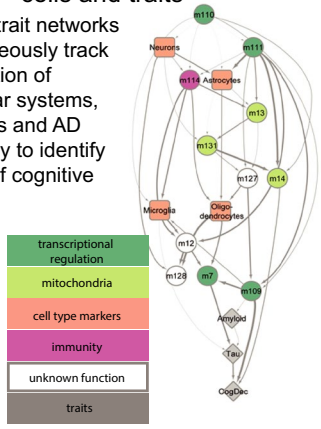
(d) Genetics and expression to identify drivers + experimental validation



This molecular network structure is predicted from genetics and gene expression data from brain samples of hundreds of individuals at various stages of AD. Circled nodes are differentially expressed in experimental perturbations of a predicted key gene identified in the network structure. Center nodes show interactions across immune components.

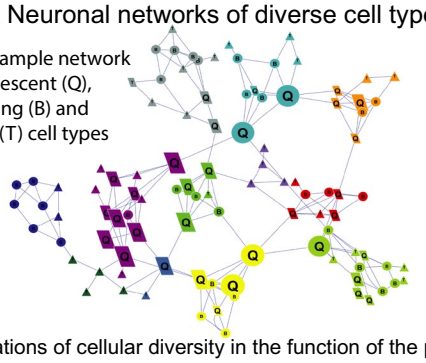
(b) Networks of molecules, cells and traits

Module-trait networks simultaneously track the function of molecular systems, cell types and AD pathology to identify drivers of cognitive decline.



(e) Neuronal networks of diverse cell types

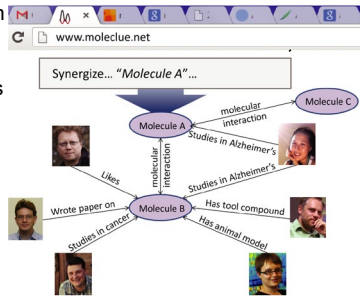
An example network of quiescent (Q), bursting (B) and tonic (T) cell types



Simulations of cellular diversity in the function of the pre-Botzinger repiratory nucleus demonstrate how strategic connectivity positions in the network influence overall activity - an interation of topology and cellular dynamics. Such effects cannot be observed without scaling electrophysiology to the level of cellular networks.

(c) Collaborator search engine using biophysics - moleclue.net

A new search algorithm to suggests scientific collaborators based on biophysical interactions among their topics of interest. This is less insular than typical influences on collboration, such as disease areas or departments, and could lead to more rational and dynamic repurposing of results.



(f) Epigenetics & mRNA to brain imaging

ROSMAP cohort combines neuroimaging and brain-based omics in the same set of hundreds of brains. These show associations between hundreds of genes and methylation of thousands of loci and brain microstructure. Shown in red is area associated with average expression of an example molecular system that is associated with cognitive decline. Green is associated with infarcts and blue with AD pathology.

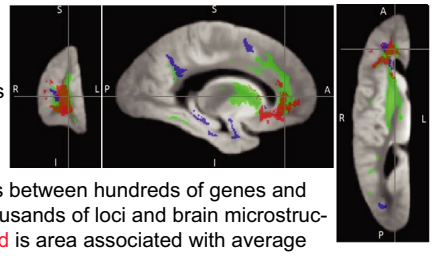


Figure 1 Multiscale modeling. Typically, scientific and drug discovery projects focus on a single biophysical scale (central column of colored nodes). The connections across these scales are obscured by the complexity of biological systems, which is an obstacle to building coherent models of disease. Coupling biophysical scales (color vertical bars between scales) allow the "scaling up" of molecular findings to the level of cognitive processes. (**a**) Combining genetics, expression, and mRNA.[136] (**b**) Considering entire molecular systems to be networks of nodes and examining their relationships to each other and disease and cognitive phenotypes. (**c**) Molecular relationships have the potential to update traditional academic relationships by identifying novel overlaps across disease areas. (**d**) One of the first multiscale systems biology approaches of combining genetics and expression information has been used to identify putative drivers of molecular networks that are associated with AD and subsequently test experimentally. (**e**) The interaction of intrinsic cellular dynamics and network topology can radically alter the output of biological systems. (**f**) Neuroimaging and molecular biology have few interactions, but with both types of data available on the same set of brains, strong, multiscale, disease-relevant coupling has emerged. AD, Alzheimer's disease; eQTL, expression quantitative trait loci; eQTH, expression quantitative trait haplotypes; eQTM, expression quantitative trait methylation; hQTL, histone quantitative trait loci; mQTL, methylation quantitative trait loci; mRNA, messenger RNA; ROSMAP, religuous order study and memory and ageing project; SNP, single nucleotide polymorphism.

scales of data, increasing the accuracy of the final predicted output. For instance, integrating genetic variants and epigenetics is useful in understanding gene regulation and the causal relationships between mRNA and disease phenotypes (**Figure 1a**).[31] Ultimately multiscale modeling for AD aims to unite the continuum of scales depicted, from genetics to the multiregion brain, allowing QSP to simulate quantitatively the effects of molecular perturbations on higher level phenotypes.

As an example, the first measurements of brain gene expression and DNA methylation alongside brain structure (via magnetic resonance imaging (MRI)) have implications for basic brain biology as well as for patient stratification and personalized medicine (**Figure 1f**).[32] This novel demonstration of relationships between gene expression and brain structure was presaged by findings that the spatial distribution of genes in the brain corresponds to neural networks[33–37] and the influence of genetic variants on brain structure.[38,39] In contrast to these spatial correlations, the combination of RNAseq, DNA methylation, and structural neuroimaging demonstrated that hundreds of genes covary with the brain microstructure across hundreds of individuals in the religuous order study and memory and aging project cohorts.[32] This observation has several implications for multiscale QSP modeling in AD. For instance, the molecular functions of the structure-synchronized genes offer potential mechanisms to influence brain structure by targeting one or more key genes in AD-relevant networks.[40] Alternatively, predicting the abundance of select molecular systems from noninvasive brain images will be helpful for stratifying individuals for drug trials. The reinterpretation of specific structural or functional neuroimaging in terms of the activity of specific molecular systems will make the findings more actionable from a drug discovery perspective. The combination of human connectomics with control theory[41] can possibly be of great interest to the QSP approach but is beyond the scope of this article. Broadly, integrating low-level (molecular) and high-level (cognition) phenotypes by including intermediate phenotypes in multiscale studies is helpful in understanding the basis of cognitive function and in describing components and model outputs for QSP approaches.

The wide-ranging data acquisition to support multiscale QSP modeling differs from typical disease research in two key aspects. First, this data acquisition links a starting point (typically genetic variants or molecules) with a specific end point (typically a disease state or symptoms such as cognitive decline) through a minimal set of intermediate scales. Second, the data are measured simultaneously or very closely in time from the same individuals. This facilitates the assessment of cross-scale coupling between biological systems, which is essential in tracking the effects of molecular (i.e., drug) interventions as they are translated into higher level phenotypes (i.e., cognition). Conversely, with data acquired in separate sets of individuals—as is typically the case—it is difficult to track the causal flow of information across multiple scales with the same degree of confidence.

In contrast to general QSP,[3] CNS QSP involves long-range neuroelectric and neurochemical signaling, feedback, and control. Knowledge gaps still exist (i) between molecular levels and electrophysiological activity, (ii) between electrophysiological activity of an individual cell and that of neuronal networks and the whole brain,[42] and (iii) how gene regulatory networks can be affected by the metabolome, either through events such as phosphorylation of transcription factors or epigenetic modulation (see also the Pharmaco-Metabolomics section). To bridge these gaps in scale, the simultaneous use of stem cell and other cellular models from the longitudinal cohort of religuous order study and memory and ageing project participants, deep learning, and massive brain simulations to scale up molecular effects to the level of cognition endophenotypes can be pursued. However, this project is likely to require at least hundreds of cell lines.

Obviously, genetic studies provide strong evidence for the involvement of nonneuronal cells, such as microglia in AD. Therefore, a valid QSP model needs to incorporate these biological processes, for instance, based on the cross-talk between microglia-derived TNF-α and neuronal activity.[43]

QSP in psychiatry

The application of advanced computational modeling in psychiatric disorders historically started with Bayesian top-down probabilistic models based on temporal difference learning.[15] This model was based on the interaction between abstract agents in the reward pathway and could explain a number of behavioral and imaging outcomes.[44] Various parameters of this agent-based approach were found to be sensitive to therapeutic interventions or brain disorders, for example, in major depression.[45] More mechanistic models based on modeling the interaction between different brain regions with a coarse implementation of excitatory–inhibitory balance have been developed and tested with clinical data.[46] This was further extended by taking into account detailed connectivity between various parts of the basal ganglia network[47] that allowed simulation of, for instance, the impact of dopamine modulation.

The development of more elaborate and biophysically realistic models of the basal ganglia[48] opened the path to introduction of pharmacology and pathology for supporting pharmaceutical R&D. Although the obvious indications were Parkinson's and Huntington's diseases,[49,50] recent developments include QSP models for psychotic symptoms.[51] Computational models for motor symptoms (including antipsychotic-drug-induced extrapyramidal parkinsonian symptoms) focused on the calculation of local field potentials in the subthalamic nucleus that could readily be tested using deep-brain recordings.[52] In contrast, a measure of the information bandwidth in the thalamic reticular nucleus was strongly associated with the clinical experience of hallucinations and delusions. As an example of late-stage (postmarketing) applications of QSP in psychiatry, optimal switching scenarios from oral antipsychotic medication to long-acting injectables—where both compounds are present in varying concentrations during the transition period—were derived.[53] Recently, mechanism-driven computational models of excitation–inhibition balance that were combined with large imaging data sets[54] suggest that cognitive deficits in schizophrenia are related to cortical disinhibition.[55]

These mechanistic QPS models can be thoroughly tested for predictability in clinical psychiatric readouts. As an example, a mechanism-based QSP model was challenged[56] to predict extrapyramidal parkinsonian motor

side-effects in clinical practice using the clinical records interactive search database[57] for schizophrenia patients in greater London treated with a combination of two antipsychotic drugs. Solely based on drug names and doses (599 unique combinations in 832 patients), in the complete absence of a training set, and just based on neuropharmacology first principles, the platform reached a modest but significant area under curve of 0.65 in a receiver operating characteristic analysis,[56] far superior to simpler predictions such as chlorpromazine equivalents. These and other achievements led to the development of a new discipline called computational psychiatry.[58]

PRACTICAL APPLICATIONS OF QSP IN CNS DRUG DISCOVERY AND DEVELOPMENT

QSP can play an important role at many different points along the trajectory of a typical CNS R&D program as illustrated in **Figure 2**. **Table 1** lists a number of examples, and we elaborate on a few interventions in the following sections.

Use of QSP in phenotypical drug discovery

The vast majority of high-throughput screens for drugs are target driven, wherein a specific molecular target is identified from its hypothesized role in a disease and cells

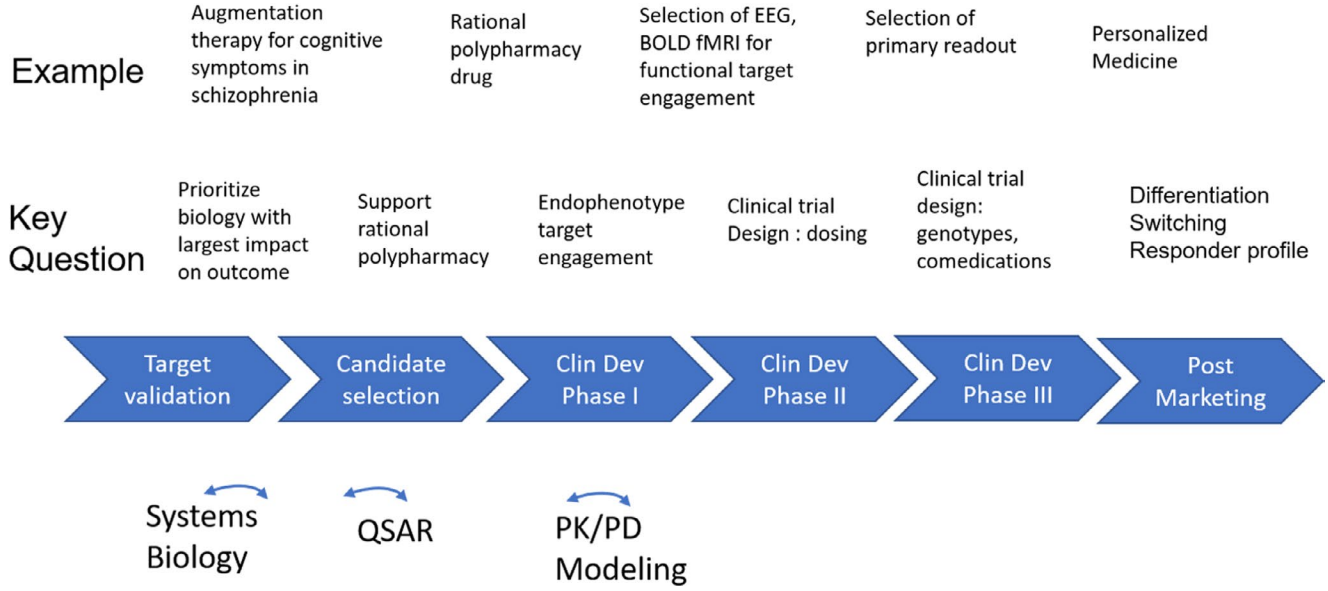


Figure 2 Impact of quantitative systems pharmacology (QSP) along a central nervous system (CNS) research and development project. Schematic overview of the impact of computational QSP along the trajectory of a CNS research and development project. QSP can help validate targets identified by systems biology studies, support rational polypharmacy and medicinal chemistry projects in combination with QSAR modeling, and better design clinical trials by predicting clinical efficacy together with PK/PD modeling. In later clinical studies QSP can model individual virtual patients to estimate the impact of comedications, genotypes and disease state on clinical outcomes. Several examples for different CNS indications are listed. BOLD, blood-oxygen level dependent; Clin, clinical; Dev, development; EEG, electro-encephalography; fMRI, functional MRI; PK/PD, pharmacokinetic/pharmacodynamic; QSAR, quantitative structure-activity relationship.

Table 1 List of major impacts of QSP on key decision points in the development of a CNS drug and comparison with traditional research and development

Decision point	Current approach	QSP
Target selection	Use of clinical–genetic data, and preclinical information	Target(s) identification with biggest impact on network and circuit outcome
Single vs. multitarget profile	Usually single target based on mostly genetic and biological information	Combination of targets based on biological information
Clinical candidate selection	Usually highly selective (avoiding side-effects)	Can be rationally designed multitarget drug or drug combination
Proof of concept dose Selection	Allometric calculations combined with *in vitro–in vivo* modeling	Can identify optimal dose in nonlinear dose response
Impact of comedication on clinical outcome	Tested when applicable	Effect predicted based on non-linear interactions between medications
Impact of genotypes on clinical outcome	Tested when applicable	Effect predicted based on nonlinear interactions with physiological effect from human imaging studies
Analysis of clinical trials	Statistical post hoc analysis; data "binning" needed for statistical power	Virtual patient analysis taking into account individual patient profile

QSP, quantitative systems pharmacology.

containing that target or an *in vitro* system using purified target are exposed to a compound library, which may include a million or more compounds, to screen for activity against the target. Once lead compounds are identified, the screening set is narrowed or refined by medicinal chemists.

In contrast, phenotypic approaches are used when researchers have not identified a specific drug target but, rather, seek to alter the phenotypic state of a disease-relevant cell, tissue, or organism. Phenotypic screens are able to target multifactorial diseases, are unbiased with regard to mechanism, and have the potential to uncover new biology, thereby creating new, best-in-class drug paradigms.[59] Numerous CNS drugs, including those that treat epilepsy and psychiatric disorders, were identified using phenotypic approaches. Historically, these compounds were the result of testing in animal behavioral models, exemplified by the discovery of antiepileptic drugs (i.e., phenytoin) that protected against "electrical seizures" in cats in 1930.[60] These phenotypic screens are limited by the facts that success relies heavily on the relevance of the disease model,[61] the target(s) may never be found nor the mechanism of action (MoA) understood, and both the screening and target identification activities are time consuming and expensive.

Modern phenotypic drug discovery relies on cell-based assays and high-throughput screening with large compound sets, although smaller but highly focused sets are used when the phenotype requires a very complex cellular model.[62] There are three main components to any phenotypic program: Generation of chemical matter from a relevant assay, studies to identify target and MoA, and finally, validation of target hypotheses. A key success factor is the "chain of translatability" from the primary assay to the disease[63] based on a shared mechanistic basis between the assay, the animal disease model, and the human disease.

As an example, spinal muscular atrophy is a disease where highly translatable phenotypic screens led to clinical candidates in part because the molecular pathological phenotype was well understood. Spinal muscular atrophy is an autosomal recessive motor neuron degeneration disease caused by the mutation or deletion of the survival of motor neuron-1 gene and reliance on survival of motor neuron-2 (*SMN2*), a paralog gene producing reduced levels of full-length protein as a result of inefficient splicing. Disease severity correlates with the copy number of the *SMN2* gene and an animal model (the Δ7 mouse) provides a clear link between the phenotypic screen and the patients. Splice modulators[64–66] discovered in the cellular systems that increase survival of motor neuron protein levels were found to be highly efficacious in the mouse model and are currently in clinical trials (ClinicalTrials.gov identifiers NCT02913482, NCT02268552).

QSP can validate the translatability of a cell-based assay to the human disease by integrating genetic and molecular data. One approach is molecular phenotyping, where the expression of a set of genes is used to infer the activation of signaling networks. A recent paper on diabetic cardiomyopathy used quantitative mRNA sequencing of pathway reporter genes to validate that their high-content imaging phenotypic assay translated to the disease tissue pathology.[67] Interestingly, this molecular phenotyping allowed clustering of their compounds and ruling out from the primary phenotypic screen false positives that did not replicate the correct molecular phenotype. Taking this a step further, a network screening approach has been proposed as a primary screen, where a minimal gene reporter set specific for a given disease is built from multiscale data sets.[68] A promising new approach, promoted by the Defense Advanced Research Projects Agency (DARPA) and still in its infancy, is to use time-resolved, untargeted multi-omic measurements to infer a drug MoA, which in its first application presented a putative resistance mechanism for cisplatin.[69]

Identifying the target or pathways involved is highly desirable but not required by regulatory agencies, as efficacy and safety are sufficient to approve a drug. However, such knowledge can greatly aid in establishing a biomarker of target engagement to help determine compound dosing, guide preclinical toxicology studies, and provide the opportunity to pivot to a target-based discovery program. Some of the tools include chemical proteomics, functional genomics, molecular profiling, and chemical informatics approaches, although no one single approach works across all situations.[70] One approach commonly taken is to screen a set of bioactive compounds that have been selected by the QSP analysis (for instance the molecular phenotyping) to gain insights into the biological processes driving the assay phenotypes and to aid in target deconvolution. If a quality chemical probe is lacking, functional genomic screens using clustered regularly interspaced short palindromic repeats/CRISPR associated protein 9, either by nuclease gene knockout or CRISPRi/a (CRISPR/i, clustered regularly interspaced short palindromic repeats/interference; CRISPR/a, clustered regularly interspaced short palindromic repeats/amplification) transcriptional regulation, can be used to identify potential targets or pathways.[71]

QSP in human-induced pluripotent stem cells

One challenge is how to incorporate the genetic basis of the various abnormal neural activities in CNS disease. Experimental disease-phenotypic neurons from a patient's human induced pluripotent stem cells (hiPSCs) are useful to the degree that cellular electrophysiological activity is related to the cognitive status of the individual at time of death and can be scaled by QSP modeling to more clinically relevant readouts through simulations of complex firing cellular networks. Such a computational platform can further "extrapolate" the experimental findings from the artificial experimental system to a more comprehensive "*in vivo*" and pathological "*in silico*" models, where additional voltage-gated or ligand-gated ion channels and network connections can be added to better simulate the human pathological condition. For instance, changes in cholinergic tone and loss of synapses and neurons can be added as in AD.[72] Because it is unclear which aspects of network firing activity (i.e., action potential width, bursting, mean activity, interspike statistics, oscillations) are most important for cognitive function, traditional classifiers or deep-learning approaches could be used to predict what property of simulated activity is associated with human cognition. A similar approach has been followed in the Comprehensive *in Vitro* ProArrythmia Assay (CiPA)[73]

or Consortium for Safety Assessment using hiPSCs,[74] with the objective to develop a predictive model for cardiac safety based on electrophysiological data with human iPSC differentiated into cardiomyocytes.

Significant limitations of this approach are the degree to which induced pluripotent stem cell (iPSC) and other cellular activity reflects that of the donor brain immediately prior to death. Similarity between the brain and derived cultures has been demonstrated to an extent for some brain diseases,[75–79] but the relationship between neural activity in a dish and human brain function remains uncertain. This potential correspondence may be enhanced by transdifferentiation of ectoderm (skin fibroblasts) and recent approaches to identify neuronal progenitors in meninges, both of which should preserve epigenetic information found in aged individuals that is lost in the generation of iPSC.[80] Another issue is whether molecular changes associated with sporadic AD will actually produce electrophysiological changes in iPSC-derived neurons and neuronal-glial cocultures. If there is a convergent signal, it could be refined or traced back to specific proteins for more rapid screening. Conversely, it is possible that convergence may be observed at the level of cellular network activity by efficiently scaling up electrophysiological simulations to the level of whole-brain activity.[81,82] Previously such person-specific brain models would be very difficult to validate; however, with the acquisition of functional MRI (fMRI) data prior and proximate to death, as is being done in the religuous order study and memory and ageing project cohorts, it is at least possible to compare the predicted blood-oxygen level dependent fMRI model (from neuronal mass models) output to the activity of the donor's brain while alive in addition to the trajectory of cognitive decline. A good multiscale QSP model can start to address the many unknowns and potential limitations to scaling molecular effects toward a cognitive readout.

An example of target identification deals with multielectrode array experiments on primary or hiPSC-derived neuronal cells to detect the effects of tau mutants on action potential propagation properties.[83] These processes can lead to altered synchronization of neuronal circuit function with important consequences for clinical readouts. QSP can simulate action potentials using a generic, computational, multicompartment neuron model with a morphology similar to the experimental setup. By performing a systematic optimization analysis, the *in silico* QSP model can reveal and prioritize molecular processes related to neuronal cell activity, such as voltage-gated and ligand-gated ion channels that reproduce the experimentally observed phenotypic changes. Further pharmacological or knock-out experiments on the hiPSC neuronal cell system can then confirm which of these processes are crucially driving the pathological changes, underscoring the bidirectional interaction between modeling and experiments.

Application of QSP modeling to clinical trials

Similar to the concept of physiology-based PK modeling where explicit biological processes are implemented to simulate PK disposition of active pharmaceutical entities, a QSP model could be envisioned as physiology-based PD modeling where the effect of interventions on biological processes is explicitly modeled on PD readouts such as biomarkers and clinical scales. Unlike traditional PK/PD modeling, these approaches enable the inference of the causal sequence and quantitative role of precise biological processes to generate "virtual patient" models to account for the variability in patient populations.

Importantly, many patients are on a multitude of brain-penetrant comedications with CNS activity that interact in complex PD ways with the drug under investigation, based on how they affect neuronal circuits. For example, in psychiatry, many patients are treated with antipsychotics with a very rich pharmacology and therefore each antipsychotic can substantially and differentially modulate the dose response of new candidate drugs in augmentation trials. Because the QSP approach is based on the implementation of pharmacology, target engagement, and neurophysiology, it is ideally suited for the simulation of these PD interactions. One QSP example involves the complex interaction of cholinomimetic drugs such as acetylcholinesterase inhibitors (donepezil and galantamine) and memantine when added to antipsychotics.[84] The effect on cognition is dependent on smoking status and the nature and dose of the antipsychotic. The number of possible combinations far exceeds the number of available patients, therefore limiting the generalizability of predictions based on data-driven "training sets." In addition, time-dependent engagement of biological targets by actual drugs in patient populations can be implemented by coupling PK profiles to the pharmacology of therapeutic interventions.

In some cases with common genetic variants, positron emission tomography imaging studies reveal the effect on brain dynamics, as is the case with the catechol-O-methyl transferase (COMT)Val156Met variant that affects enzyme stability and dopamine dynamics[85] or the Serotonin Transporter rs 23351 s/L genotype affecting the expression of the serotonin transporter in the human brain.[86] These genotypes can significantly affect cognition and mood.

A mechanism-based CNS QSP model can incorporate the pharmacology of many approved CNS active medications, together with their level of functional intrasynaptic concentration,[87] whereas the effect of common variants or smoking on neurotransmitter dynamics can be derived from positron emission tomography imaging studies in unmedicated healthy volunteers.[85,86,88] This in turn allows the prediction of the dose responses of a new investigative drug in the presence of these different PD-PD interactions. Failure to account for these PD-PD interactions in a clinical trial can lead to imbalance of allocation between treatment arms and reduce the clinical signal, possibly leading to false negatives. In contrast, traditional statistical *post hoc* analysis of a clinical trial often fails to take into account the individual nature of each patient, as patients are often grouped together to achieve statistical power. Mechanism-based QSP "virtual patient" models can identify these possible problems before starting clinical trials.

Application of systems pharmacology approaches to safety assessment

With the advances in informatics and availability of big data, systems approaches can facilitate drug development by integrating the molecular hierarchy of human biology related

to a drug candidate's action with specific clinical phenotypes of the target disease.[89] Using publicly accessible high-quality databases and knowledge bases of mRNAs, genomics, and protein interaction networks, systems pharmacology modeling has identified the biological factors that underlie drug-induced peripheral neuropathy[90] or that are commonly shared by drug-induced rhabdomyolysis, cardiomyopathy, peripheral neuropathy, Steven-Johnson syndrome, and lung toxicity.[91] Recently, potential molecular biomarkers for drug-induced cardiomyopathy have been identified using transcriptomic data from human cardiomyocytes.[92] When considering CNS side-effects, prediction of seizure, or abuse liability, sedation and cognitive impairment come to mind. Recently, microelectrode arrays–based technology has been used in combination with physiology-based PK modeling to predict seizures in primary cultures of rat cortical neurons.[93] In another example, three human cell lines were characterized as models for high-throughput neuronal cytotoxicity screening.[94] These systems pharmacology modeling efforts lay the foundation for QSP-based predictive assessment of specific dose-dependent CNS toxicity, therefore accelerating precision medicine and reducing drug attrition.

The evaluation of systems pharmacology/mechanistic models have been part of the US Food and Drug Administration (FDA)'s model-informed drug development pilot program.[95] In addition to demonstration of target engagement and support of preclinical drug discovery and development, QSP/mechanistic models have been applied to various stages of clinical drug development.[96] Recently, QSP modeling and simulation provided supportive evidence for the approval of a new dosing regimen. The FDA strives to engage in efforts with stakeholders to use innovative quantitative approaches, such as QSP to guide development of safe and effective drugs to treat diseases including those of CNS.

NEW DEVELOPMENTS IN CNS "SYSTEMS" APPROACHES

The integration of wearables to acquire disease-progression data

It is clear from the previous discussions that each patient is unique and that in principle mechanism-based QSP approaches can address this variability. Having access to large databases with individual outcomes is mandatory for the validation of these models. Sage Bionetwork's mPower Progression research study[97] provides an example of how cell phones can be used to expand the definition of the system under study and gather new individual patient data on Parkinson's disease (a half million activities were recorded from 9,520 unique participants over 19 months). At enrollment, the participants answer a brief questionnaire and download a cell phone app for daily, participatory measurements to track symptoms, triggers, and medications; measure gait and balance; evaluate tapping speed, coordination, and tremor; and assess memory and thinking. A QSP platform, informed by these data, should promote an improved understanding of the biology behind Parkinson's symptoms, identify druggable targets for patient subgroups, and lead to the validation of a digital biomarker for diagnosis and treatment. One strategy would consist of focusing on modeling the biology of the most relevant pathway for a specific symptom based on domain expertise and including the effect of medications and other factors that affect this outcome and ultimately constraining the QSP model using individual patient data.

Large-scale collaborations targeted to CNS diseases

Coordinated integration and public dissemination of data and evidence across these projects provides a powerful way to increase the reproducibility and translatability of discovery research, enabling rapid advancement of systems pharmacology concepts in AD drug development.

For instance, a number of research entities, clinic sites, multiple sponsors, and a large population of ALS patients comprise the Answer ALS consortium,[98] aiming to collect omics data from motor neurons derived from iPSCs obtained from 1,000 patients, with the goal of extracting underlying causal molecular networks and disease subtypes for this devastating disease. Answer ALS is also collecting and curating proteomic, epigenetic, whole-genome sequencing, metabolomics, and RNAseq data, with matched clinical information. The integration of these data requires the use of models that are sufficiently flexible to incorporate various types of data and can leverage statistical information. A first example is the crowdsourcing effort to develop predictors of clinical progression in ALS.[99] In a complementary way, mechanism based QSP is also able to account for underpowered, data-poor realms by integrating formalized domain expertise and can generalize beyond the original patient population.

A related example and resource is the Library of Integrated Network-Based Cellular Signatures (LINCS) program,[100] supported by the NIH Common Fund. LINCS is a multicenter effort to catalog how human cells respond globally to chemical, genetic, and microenvironmental perturbations and disease-causing mutations.[101] The measurements include but are not limited to transcript profiling, mass spectrometry, cell imaging, and biochemical techniques (**Box 3**). These data can inform mechanism-based computational QSP to generate predictive models of regulatory and signaling networks.[102]

New *in vitro* model systems

Although vast sums have been invested with the goal of understanding the brain and its disorders, much of this work has been conducted using animal and *in vitro* models that are not sufficiently relevant to lead to cures or treatment of complex human disorders.

One approach is to minimize the distance required for *in vitro* to *in vivo* extrapolation. *In vitro* tissue-engineered models that use human CNS neurons, support cells, and vasculature enable live cell imaging and quantitative analysis of aspects of gene expression and drug delivery to the brain that are difficult to obtain from studies in animal models. These human microphysiological systems (MPS) are two-dimensional and three-dimensional cellular constructs that can be used alone to recreate the function of single organs or coupled together into organ systems and even multiorgan MPS "homunculi-on-a-chip." The following three general areas must be

optimized: (i) the sourcing of well-characterized human cells from healthy and diseased patients, (ii) the design of microbioreactor systems that support engineered organoids, tissue constructs, and biological interfaces, and (iii) the analytical tools and techniques required to record and analyze data from both the *in vitro* models and *in vivo* humans.

Many physiological functions and diseases are the product of a complex tissue microenvironment that may be controlled by tissue interfaces, with the best example being the neurovascular unit (NVU),[103,104] which encompasses not only the BBB but also the brain's heterogeneous population of neuronal and supporting cells. Simple microfluidic NVUs already demonstrate the modulation of BBB permeability with drugs and inflammatory cytokines.[104] Real-time imaging of drug transport can be used to develop quantitative kinetic models of transendothelial transport that link the PK and PD of drug and gene delivery, particularly with regard to efflux transporters that can be upregulated in certain conditions. MPS models enable the real-time observation of synapse formation when adjacent populations are brought into chemical contact.[105–107] Ultimately, the NVU will have a self-organized, hierarchical vasculature and BBB and will inform the development of computational QSP *in vitro* to *in vivo* extrapolation.[108,109]

MPS models of the BBB allow observation of a cross-section of the BBB[110] and the binding of nanoparticle drug carriers to the brain endothelium,[111] organophosphate toxicity,[112] and the effects of methamphetamine on the BBB and metabolic coupling of the BBB and neurons.[113] Other examples include a human brain spheroid model that was recently validated for neurodevelopmental toxicity[114] and for drug development and potentially for personalized medicine.[115] A major challenge is to couple models of processes of metabolism that are governed by the distributed properties of complex organ systems, for example, the gut-liver-brain axis.

Biomarker discovery and validation

There is an urgent need for validated biomarkers that could be used to establish diagnosis, guide drug selection, and reliably predict individual variation in response to treatment among patient subpopulations.[116] Recently, large cohorts of patients with deep phenotyping are identifying new biomarkers in relation to individual clinical trajectories. Although CNS pharmacology has yet to fully exploit the massive amounts of -omics data, the field is now poised to benefit from the incorporation of quantitative techniques that can integrate these massive amounts of data to generate actionable knowledge.

New approaches in statistical learning can support the identification of biomarkers from high-dimensional data, as exemplified by the use of machine learning to combine molecular, imaging, behavioral, genetic, transcriptomic, demographic, cognitive, and environmental data to identify biomarkers and biosignatures for substance abuse disorders such as nicotine addiction.[117] Traditional statistical approaches for identification of simple molecular biomarkers from large, generally homogenous data sets are ill suited for the specification of multimodal biomarkers for which the number of factors being tracked can vastly exceed the number of individuals in the study. Machine learning, other artificial intelligence algorithms, optimal design of experiments, and mathematical techniques for dimensionality reduction will prove invaluable in identifying the biomarkers.[118] However, the findings of such approaches are as good as the quality of the training set data, which are often less than ideal in clinical situations. In many cases, these approaches yield correlations and associations but not necessarily causal relationships A mechanism-based QSP model can partially uncover causality by integrating formalized domain expertise on the pathways or circuits identified in these analyses, as proposed previously.[119] Once these challenges are resolved, insights from the integration of QSP simulation with these -omics databases would advance the development of biomarkers in association with a mechanistic understanding of the pathways involved for both the clinical and regulatory environments.

Pharmaco-metabolomics

Recently, the two-way interaction between the gastrointestinal microbiome and the CNS, known as the gut–brain axis,[120–123] has been recognized as playing a key role in health and disease, for instance in Parkinson's disease.[124] Human biology is affected by external factors that include drugs, lifestyle, environmental toxins, age, and social factors possibly through a two-way interaction between the gastrointestinal microbiome and the CNS. From this perspective, a patient's response to a drug can be observed by tracking changes in both the human and microbiome metabolome. Classic biochemical approaches focus tightly on single metabolites, their reactions and kinetic properties, and/or defined sets of linked (i.e., precursor/product, intermediary metabolism) reactions and cycles. In contrast, metabolomics collects quantitative data on a broader series of metabolites to gain an overall understanding of metabolism and/or metabolic shifts associated with conditions of interest.

The collaboration between the Pharmacogenomics[125] and Pharmacometabolomics[126] Research Networks explores the metabolic state of an individual as affected by environmental, genetic, and gut microbiome influences—the "metabotype"—to define biosignatures before and after drug exposure to inform treatment outcomes, providing tools for disease subclassification and identifying pathways contributing to drug–response phenotypes. This expands the wide scope of the various -omics technologies for which future data might uncover new modulating pathways for CNS disorders.

These applications can identify metabolic influences on drug PK as well as PD influences of metabolism on the target itself and downstream signaling. Ideally, metabolomics will ultimately be able to contribute a detailed map of the regulation of metabolic pathways and therefore of the interaction of proteins encoded by the genome with environmental factors, including drug exposure informing mechanism-based computational QSP approaches.

However, an important not yet fully resolved issue is the degree to which peripheral processes, measured by pharmacometabolomics, reflect the central processes in the brain.[127] As a notable example of how coupled-organ MPS

systems might contribute to this understanding, it has been recently shown *in vitro* that a microbial metabolite trimethylamine, metabolized by the liver into trimethylamine-N-oxide, can transfer across the BBB,[128] which was subsequently confirmed with *in vivo* measurements of human CSF.[129] Future QSP approaches should take such findings in consideration.

Another important application of the same technological advances is the quantitative measurement of posttranslational modifications, such as truncation, phosphorylation, and acetylation affect in exquisite detail the functionality of key proteins such as amyloid and tau in AD.[130] These changes, either from postmortem studies or detected in CSF or plasma, are beyond the reach of genomic and transcriptomic studies and add another important layer of knowledge to any QSP model.

LOOKING TO THE FUTURE

The pharmaceutical industry is primarily looking for answers to practical questions covering the whole spectrum of R&D such as target validation, translational extrapolation from preclinical animals to humans, the selection of the best clinical candidates, optimization of clinical trial design, and patient enrichment strategies. Some of these issues can be addressed by current modeling methodologies, such as PK/PD and disease modeling;[131] however, these are based on retrospectively fitting clinical data with empirical equations and therefore often fall short in projects for new as yet untested targets.

There are a number of specific challenges for a wider acceptance of CNS QSP. First, there is a lack of skilled modelers, ideally domain experts with modeling expertise. For relatively simple processes, helpful software tools have been developed; however, for more complex situations such as firing of interacting neuronal networks, the learning curve is often steep. This issue is not necessarily limited to CNS modeling; in larger organizations, modelers are shifted around depending on the priorities of the indications pursued. This hampers the acquisition of expertise in a particular disease area, which is especially crucial for the complex brain neurophysiology. Second, the "validation" of any new technology can only happen by comparison with experimental clinical data, which are often difficult to obtain and are only available much later in the project. In this context it has to be noted that animal models for AD, given all the failed clinical trials are basically not validated either. Third, the assumptions of these computational models are perceived to be largely incomplete, in contrast to animal models, but these advanced models are much more grounded in the human clinical situation, suggesting that these two approaches are complementary. Finally, the development of these complex brain disease models is likely beyond the capabilities of a single company, hence the need for precompetitive public–private consortia.

It is important to underscore the role of neuronal circuit firing because brain activity in specific regions drives clinical behavior. The effect of therapeutic interventions on biomarkers (e.g., CSF β-amyloid or tau levels) is important for target engagement; however, the primary readout demanded by regulatory agencies in clinical trials is a beneficial effect on behavioral clinical scales, such as Alzheimer's disease assessment scale cognition and clinical global index. This has been dramatically illustrated by recent clinical trials with amyloid modulating agents, where despite a significant effect on a "relevant biomarker," there was no clinically relevant benefit.[132]

A unique but less publicized application of a validated QSP approach is the capability for reverse engineering to prioritize the biological processes that are key drivers of a reversal of a pathological readout to a "healthy" readout. Using search strategies for optimization that are common in engineering sciences, it is possible to prioritize the biological processes that are candidates for therapeutic interventions. This allows the extension of the concept of "target identification," which is the purview of correlative systems biology, by adding a level of target validation. Because the platform is quantitative in nature, quantitative properties of such intervention are easily determined (in what direction and how much the pathway needs to be changed?). Finally, it allows for the identification of synergy between different therapeutic interventions, laying the groundwork for a rational polypharmacy guided medicinal campaign.[133]

Partnerships to facilitate the optimization of QSP in CNS drug discovery

The AMP-AD[134] and the Molecular Mechanisms of the Vascular Etiology of Alzheimer's Disease (M^2OVE-AD)[134] consortia are sponsored by the NIA for systems biology approaches to quantify network-based human AD states in support of target prioritization and biomarker discovery. The Model Organism Development and Evaluation for Late Onset Alzheimer's Disease (MODEL-AD) consortium generates rodent models of late-onset AD based on genetic and genomic observations, including those arising from AMP-AD and M^2OVE-AD. This collaboration provides shared resources, formalized evaluations of methods and outputs, interactive collaborations, and opportunities to educate and translate findings across researchers with complementary but diverse expertise. As a result of the need for longitudinal data sources for cognitive measures associated with postmortem brains, the process requires sustained commitment. As many pathologies contribute to dementia,[135] hundreds of brains are needed to produce stable modeling results.

The future of QSP

It is crucial to emphasize that computational QSP (i) is based on formalizing existing knowledge; (ii) simulates known and well documented physiological pathways; (iii) is quantitative and causative rather than qualitative and correlative; (iv) has a focus on pharmacological interventions, including new untested targets; and (v) can have its biological assumptions interrogated and challenged, in contrast to the "black box" of machine learning.

Recommendation 1: Emphasize the importance of computational modeling in the training and continuing education of neuroscientists. The skills set for developing computational QSP models in CNS is usually not in the curriculum of bio-informaticians and often present a steep learning curve because a neurobiological understanding

or domain expertise is mandatory to develop a superior predictive model. In our experience, the best CNS QSP modelers start out as bona fide neuroscientists who learn a modeling platform rather than the other way around. They are also best positioned to help interpret the outcomes of the simulations that are often unexpected because of the many nonlinear interactions.

Recommendation 2: Integrate QSP in the whole R&D process. Another issue is the lack of concrete short-term return on investment for introducing QSP as a standard operating procedure. Because it is mechanism based, it often takes substantial time and effort to develop such complex models, and the major outcome is often a better understanding of the biological processes involved, which at the stage of early discovery might still require many years to become useful. In this article, we argued that computational QSP can be helpful at different stages of the development cycle with possible short-term returns such as the impact of QSP on clinical trial design or analysis of failed trials at the single patient level.

Recommendation 3: Develop QSP in a precompetitive consortium. Similarly, the same computational complexity might make simple fit-for-purpose models less interesting; instead we see this approach more as an ever-evolving platform that captures increasing levels of detail across a widening range of scales. In that sense, a possible precompetitive consortium in an open-access research center where complex CNS QSP models would be developed and where every partner has access to these models for their own internal projects is a possible solution that could also solve the relative scarceness of specific expertise.

Recommendation 4: Consider QSP as a tool for human augmented intelligence. By definition, any QSP model is incomplete; often it does not include all the detailed processes that scientists are working on, and often these models are judged by the lack of these details rather than the overall emergent properties that are closer to the clinical readout. Because of the transparency of the assumptions, the QSP model can be expanded by incorporating different hypotheses proposed by the scientists and calculating the impact at the level of complex neuronal circuit properties. Therefore, we see the QSP approach as a tool for augmented human intelligence; it supports the human mind to better grasp the complexity of brain disorders. This is especially true in understanding the biological basis of the many clinical trial failures so that going forward the probability of successful clinical development can be increased.

Recommendation 5: Develop systematic high-quality knowledge map. Similar to machine learning, the quality of information that drives the development of QSP is of utmost importance. Instead of relying on a patchwork of expert knowledge of varying quality, breadth, and depth, an idea is to build a systematic evidence map of existing literature of the CNS disorders, incorporating and linking together multiscale human and other mechanistic and personalized patient and temporal information. Such an evidence map will identify the knowledge gaps and help formulate specific questions in areas where there is enough published literature, possibly compiled into in a series of systematic reviews. The systematic review is a framework and a methodology that is well established in clinical trials and spearheaded by the Cochrane Collaboration (https://www.cochrane.org) for answering well-framed specific questions. The systematic reviews are considered the top level of evidence in clinical medicine for their comprehensiveness, transparency, and objectivity. Although not a small undertaking, this approach, now a foundation of evidence-based medicine, will pay off to become a foundation for a multiscale QSP model, where one could manipulate various endpoints to observe changes in predicted phenotypes, test drug target hypotheses, and iterate on various conditions. Such foundational work should be done with input from all stakeholders and is ideally conducted in a precompetitive multistakeholder consortium. The transparency and involvement in building such a knowledge map will remove the barriers identified in this workshop and will spur innovation in CNS drug discovery, uncovering new disease pathways and targets and developing new human mechanism-based models for testing the QSP-derived approaches.

In conclusion, with all the caveats previously mentioned, computational CNS QSP is gradually evolving into a mature technology and could have the capability to move beyond the prediction of effect on biomarkers toward functional clinical responses of pharmaceutical interventions in CNS diseases.

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