Pharmacodynamic Evaluation: CNS Methodologies

Lynne Hughes, Marie Trad, Stacey Boyer, Deborah Lee, and Wei Yin

Contents

L. Hughes (✉)
Neurology Center of Excellence, IQVIA, Reading, Berkshire, UK
e-mail: lynne.hughes@iqvia.com

M. Trad
Neurology Center of Excellence, IQVIA, Paris, France
e-mail: marie.trad@iqvia.com

S. Boyer
Clinical Research Operations, Cavion, Charlottesville, VA, USA
e-mail: boyer@cavionpharma.com

D. Lee
CNS-TAU, Takeda Pharmaceutical International Co., Cambridge, MA, USA
e-mail: leezwiz@yahoo.com

W. Yin
Quantitative Clinical Pharmacology, Takeda Pharmaceuticals, Cambridge, MA, USA
e-mail: wei.yin@takeda.com

F.J. Hock, M.R. Gralinski (eds.), *Drug Discovery and Evaluation: Methods in Clinical Pharmacology*,
https://doi.org/10.1007/978-3-319-56637-5_32-1

Abstract

Drug discovery in neurology is confronted with a high attrition of compounds in both early and mid-stages of the development cycle. The causes of these failures are multiple and mainly based on the uncertainty around the precise pathological processes, lack of reliable biomarkers, and variability of drug penetration through the blood-brain barrier (BBB) into the brain. The first section of this chapter focuses on pharmacokinetic/pharmacodynamic (PK/PD) aspects when developing a medication targeting brain disorders. The biomarker section discusses those markers employed in the most common neurological disorders. It addresses their advantages and limitations, and how they serve in confirming efficacy and, in some instances, the safety of therapeutic interventions.

Pharmacokinetics and Pharmacodynamics in Neurology

Stacey Boyer, Deborah Lee, and Wei Yin

Introduction

Drug development efforts to treat neurological disorders have continuously faced significant challenges, leading to a lower success rate in comparison with other therapeutic areas (Reichel 2009). Of these difficulties, measurement and accurate prediction of the free drug concentrations at the site of action within the brain, and identification of pharmacodynamic (PD) biomarkers for the mechanism of action and drug effect are of particular focus. Since pharmacological activity depends on the free drug concentrations at the site of action, it has become critical to optimize unbound drug concentrations in the central nervous system (CNS), most notably at the drug target. The unbound drug concentrations can be measured in both in vitro and in vivo preclinical models (Reichel 2015). In patients with neurological diseases, obtaining information with regards to substance penetration into the CNS and target exposure requires application of biological sampling and translational techniques (de Lange 2013). Despite these limitations, there is opportunity to apply novel concepts and optimize existing methodologies and tools to assess drug penetration and pharmacokinetics (PK) in the human brain (Reichel 2015; Rizk et al. 2017).

Factors that Control Target Drug Exposure in the CNS

Critical factors that control the free drug exposure at the CNS target include systemic PK, plasma protein binding, rate and extent of CNS penetration, CNS distribution, binding to brain tissues, and elimination (de Lange 2013; Reichel 2015). Systemic PK and plasma protein binding determine the unbound free plasma concentration. Since only the unbound drug is able to pass through membrane barriers (e.g., the BBB), it is important to measure the unbound plasma concentration to understand drug transport to the brain (de Lange 2013). The rate of CNS penetration is controlled by cerebral blood flow and permeability across the BBB. The extent of CNS penetration is not determined by the permeability and the penetration rate: rather, it is measured by the unbound brain to unbound plasma concentration ratio (Reichel 2015). CNS distribution is also the key element in determining the effective concentration at the CNS target. Drug elimination from the CNS via extracellular fluid (ECF) bulk flow into the cerebrospinal fluid (CSF) or across the BBB reduces drug concentration at the target site (de Lange 2013; Reichel 2015). Drug metabolism

at the BBB and blood-CSF barrier may also influence CNS distribution accordingly (de Lange 2013).

Methods to Evaluate Brain Penetration and PK in the CNS

Evaluation of brain penetration and PK in the CNS is conducted via in vitro and in vivo studies, in situ brain perfusion, and integrated quantitative modeling approaches (Alavijeh et al. 2005; Danhof et al. 2007; de Lange 2013; Reichel 2009, 2015; Rizk et al. 2017; Yamamoto et al. 2017). In vitro studies include permeability and transporter assays using Caco-2 and MDCK-MDR1 cells, and binding assays in plasma, brain homogenate, or brain slices (Reichel 2009). In situ brain perfusion in whole animals can also provide a kinetic measure of drug entry rate into the brain (Alavijeh et al. 2005). A notable in vivo study is the determination of the unbound brain/plasma ratio in rodents, which provides information on the extent of brain penetration. In addition, CSF samples are of high value since they can be collected in humans to provide information on unbound drug concentration in the CNS.

To better understand the concentrations over time in ECF, a key compartment for the effect of many drugs, brain microdialysis in rodents can be applied to calculate PK parameters such as Cmax, $t_{1/2}$, and AUC (Reichel 2009; Alavijeh et al. 2005). Physiologically-based PK (PBPK) models have also become increasingly important to distinguish between system- and drug-specific parameters to allow for translational prediction of human CNS PK and associated drug effect from preclinical models accordingly (Yamamoto et al. 2017; de Lange 2013).

Translating PK/PD to Address Neurological Disease

As discussed in further detail in due course, translational approaches to predict exposure and distribution in the human brain present both inter- and intraindividual variation when investigating therapeutic modalities targeting the CNS (de Lange 2013). Addressing these variations is required to understand PK and effects in the CNS, as they differ between diseases and species (de Lange 2013). Consideration must also be paid to the variation in PK properties of different compounds as they relate to defining dose regimens and their pharmacological and/or toxicological effects (Dingemanse et al. 1998). Both PK and PD properties of the drug under study should be well defined in terms of concentration, dose-response relationships, and target site kinetics as they relate to the translation of preclinical models to clinical study (Dingemanse et al. 1998).

Target Exposure and Target Engagement in the CNS

Both target exposure and target engagement are required to foster target-mediated pharmacology and therapeutic effect(s) on the etiology of the neurological disease (Reichel 2015). Target exposure lends itself to PK optimization efforts focused on both unbound and total drug concentrations (Reichel 2015). In the absence of minimally required exposure, the drug will be unable to mediate the necessary required pharmacology to yield the desired effect(s) on the neurological disease under study (Reichel 2015). Target engagement requires binding of the drug to the target protein at concentrations in excess of the pharmacological potency of the drug over time (Reichel 2015). Within the CNS, target site (s) can be incredibly challenging for drug access, as the BBB limits exposure as a result of tight cell junctions and multiple efflux transporters (Reichel 2015). As a result, neurological drug discovery has focused on assessing target engagement via brain tissue sampling in preclinical models of CNS diseases in humans: in addition, indirect methodologies such as imaging modalities can provide demonstration of target engagement (Durham and Blanco 2015).

Translational Approaches

Translation from preclinical models to the clinic must account for drug potency to normalize across species for differences in target receptor inhibition, PK to inform differences in dose-exposure, and level of the PK/PD relationship to facilitate prediction of CNS penetration and efficacy (Di and Kerns 2015). Limitations include

contributing factors in the drug concentration-time profile which influence PK in the CNS, and physiological volumes of CSF: intracellular fluid and interstitial fluid should also be considered when translating exposure to efficacy (Di and Kerns 2015). Integrated PK/PD modeling essentially serves as a valuable tool, merging quantitatively driven in vitro pharmacologic properties of a drug with its respective in vivo PK to investigate the exposure-response (E-R) relationship (Tuntland et al. 2014).

Determining Target Engagement in the CNS via Positron Emission Tomography

While scaling of PK properties is executed via application of standard allometric principles, PD properties are often species-independent as related to receptor occupancy (Melham 2013). A noninvasive approach to determining target engagement in the CNS is the application of positron emission tomography (PET), an imaging technique which enables translation of ex vivo receptor occupancy from preclinical models to humans (Melham 2013). Application of PET provides a quantification of brain metabolism, receptor binding of various neurotransmitter systems, and of alterations in regional blood flow (Politis and Piccini 2012). Translation of research into clinical populations may assist with differential diagnosis and narrowing of complexities in patients with neurological disorders such as mild cognitive impairment and dementia (Johnson et al. 2013).

PK/PD Relationships in Humans

The fundamental understanding of PK/PD or E-R relationships is not always straightforward since it relates to a pharmacological or toxicological response relative to the drug concentration at the site of action. The drug concentration needs to reach a minimal level to obtain a response. E-R relationships can be complex since they are not always direct and rapidly reversible (Dingemanse et al. 1998). In addition, various factors can impact both PK and E-R relationships, including age, sex, race, disease status, chronic drug use, and drug-drug interactions. As more detailed information on the PK and E-R relationships becomes available for CNS drugs, well-defined E-R relationships have been used to optimize dose regimen in neurology clinical trials. This is notably so in the late phase to increase probability of success and to extrapolate the dose regimen in special populations (e.g., elderly, pediatric) (Dingemanse et al. 1998).

PK/PD Effects of Drugs in Neurological Disorders

Approved cholinesterase inhibitors (i.e., tacrine, donepezil, rivastigmine, and galantamine) for first-line treatment for the symptoms of Alzheimer's disease (AD) patients demonstrate a dose-related effect on the desired therapeutic outcome of improved cognition and functional activities, as well as the mechanism-based gastrointestinal adverse events.

The application of antiepileptics to treat neonatal neurological diseases has been very challenging owing to the lack of reliable data to inform safe and effective dose regimens in this patient population. The majority of the drugs utilized are done so in an off-label manner, leading to scaling from adult dose based on body weight or body surface area (Donovan 2015). As a result of these challenges, quantitative modeling approaches become essential to optimize the dose selection for the neonatal population. For example, a population PBPK model simulation suggested that only 10% of the adult dose of lorazepam needs to be given to newborns to demonstrate antiepileptic activity (Donovan et al. 2015; Maharaj et al. 2013).

PK/PD data following administration of antiretrovirals indicate highly variable drug transfer to the CNS, as CNS penetration is dependent on both drug and patient characteristics (Calcagno et al. 2014). The use of drugs with high penetration and compartmental activity has been associated with optimized CSF viral control, and in some cases, enhanced neurocognitive activities (Calcagno et al. 2014).

PK/PD Modeling of Neurology Drug Effects: Utility and Implications

As discussed, developing drugs to treat neurological disorders involving the CNS is particularly challenging, often because of many factors including the complexity of brain function, relative isolation of the CNS from peripheral drug compartments, lack of definitive pathology, inability to clearly monitor the effects of drugs, lack of biomarkers, both inter- and intra-variability of drug delivery to the CNS, difficulty of prediction of appropriate dosing in alternative populations such as children, and inability to predict both alternative on- and off-target effects. In particular, the first hurdle is often ensuring that the drug reaches the target "at the right place, at the right time, and at the right concentration" (de Lange 2013).

In this section, the ability of PK/PD modeling to help predict brain distribution, kinetics, and therapeutic effects, how PK/PD models can be used to predict various effects of differing formulations over a broad dosing range, and how PK/PD extrapolation can lead to regulatory approvals will be discussed.

Accessing the CNS

The first hurdle facing drugs to treat CNS disorders is successful access to the appropriate CNS compartments, i.e., the brain extracellular fluid compartment, brain intracellular compartment, and ventricular and lumbar CSF compartments. Mechanisms of crossing the BBB and the blood-CSF barrier include passive diffusion of unbound molecules across a concentration gradient, facilitated diffusion across a concentration gradient using a helper molecule, vesicular transport, and active transport, which may occur against a concentration gradient but requires energy. Even if the drug gains access to the CNS, drug effects may be independently modified by CSF turnover and ECF bulk flow, extra-intracellular exchange and brain tissue binding, drug metabolism, target interaction, and signal transduction and homeostatic processes, all of which can vary between individuals and disease states. Other individual factors that can play a role in determining drug effect include genetic background (e.g., fast vs. slow metabolizers), sex, age, diet, and long-term drug treatment with the drug of interest or concomitant medications (i.e., up- or down-regulation of receptors). Finally, while most of the elegant and informative PK/PD studies are carried out in animals, interspecies differences are well known.

Because of these many variables, in vivo experiments need to be integrated. Data should be collected examining multiple different conditions in the same animals, employing study designs in which as many conditions that can lead to variability as possible are taken into account. These may include, but are not limited to, protein binding, inhibition of transporters and receptors, and/or existence of the disease state which closely mimics the human disease. In addition, in vivo dialysis methods in animal models may establish the effects that bound and unbound drugs may have in CNS drug delivery.

The Mastermind Approach

To help understand the complexity of the integrated studies and the data expected to result from them, de Lange (2013) proposed using the Mastermind approach. The game Mastermind is a unique code breaking/code making board game in which colored pegs are used by one player to establish a code and by the other player to systematically break the code. In a similar manner, de Lange (2013) hypothesized that the "code" represents a complex PK/PD relationship. By understanding the strict distinction between the properties of the drugs and the properties of the biological systems by careful design of integrated studies and by utilizing mathematical modeling, the "code" can be broken. De Lange (2013) uses this Mastermind approach and detailed PBPK modeling to understand drug effects of acetaminophen, quinidine, and remoxipride.

Developing PK/PD Models from Human Data to Inform Future Clinical Trials

While de Lange (2013) proposed detailed integrated animal studies and mathematical modeling to predict CNS effects in humans, studies in humans can also be used to build PK/PD models.

These models can then be employed to predict time of onset and expected effects of alternative dosing regimens not tested. In one example, Wiltshire et al. (2012) reported such a model based on a Phase 1 single-ascending-dose study of remimazolam, a rapidly metabolized benzodiazepine when compared with placebo and midazolam. In this randomized, single dose-escalation study, 54 healthy subjects in 9 groups received a single 1-min IV infusion of remimazolam (0.01–0.3 mg/kg), while 18 subjects received midazolam or placebo. All data were used for Monte-Carlo simulations of alternative dosing regimens. A 4-compartment pharmacokinetic model of midazolam and a physiologically based recirculation model of remimazolam best fit the data. Simulations based on these models demonstrated that remimazolam delivered extremely rapid sedation and that dosing by body weight offered no advantage over the weight range studies (65–90 kg) suggesting a fixed dose regimen.

Sophisticated PK/PD modeling has been accepted by regulatory agencies for extrapolation of efficacy from one population to another. In 2017, the International Conference on Harmonisation (ICH) revised a guideline that states as follows: "When a medicinal product is to be used in the pediatric population for the same indication(s) as those studied and approved in adults, the disease process is similar in adults and pediatric patients, and the outcome of therapy is likely to be comparable, extrapolation from adult efficacy data may be appropriate." (ICH 2017). In 2012, Pellock et al. demonstrated that antiepileptic drugs from published trials in focal seizures showed clinical responses that were similar in both children and adults based on comparable PD effect sizes. Based on this, the Pediatric Epilepsy Academic Consortium on Extrapolation (PEACE) was formed. After clear demonstration using both animal models and information in humans that the physiological processes which lead to focal seizures in adults are similar to those in children as young as 2 years of age, there was regulatory agreement that sophisticated PK/PD models could be used to extrapolate efficacy from the adult population to the pediatric population (Pellock et al. 2017). However, it was recognized that enough PK data needed to be obtained from pediatric populations to determine actual exposure which then can be used to predict efficacy based on PK/PD models, and that extrapolation could not be used to determine safety.

A novel use of PK/PD modeling led to approval of vigabatrin for pediatric patients with partial seizures (Nielson et al. 2014). Previously, vigabatrin had been approved for pediatric patients between 1 month to 2 years of age with infantile spasms, and as adjunctive therapy in adult patients with complex partial seizures who had responded inadequately to several alternative treatments. Upon approval, the FDA issued a Pediatric Research Equity (PREA) requirement for children with refractory partial seizures. Three previous Phase 3 trials had been initiated but were halted early for administration reasons. These data were pooled with adult pivotal clinical trial data to develop a population dose-response model linking vigabatrin dosage and seizure counts. This model allowed prediction of appropriate dosing based on body weight. Submission of these data along with the model simulations allowed for the successful fulfillment of the PREA requirement and approval by FDA of vigabatrin in pediatric patients with complex partial seizures.

Summary

Discussions to this point in the chapter demonstrate that while more complicated than other systems, PK/PD modeling can be successfully utilized to better understand the significant complications involved in achieving CNS drug delivery to the correct location at the correct time and with the correct concentration. Data obtained in limited clinical studies can be expanded using these models to predict the full extent of dosing effects and even lead to regulatory approval. Attention now turns to biomarkers.

Biomarkers in Neurology

Lynne Hughes and Marie Trad

Introduction

With the increased global life expectancy, the prevalence of neurodegenerative diseases such as Alzheimer's disease (AD) and Parkinson's disease (PD) is expected to grow significantly in the coming years. The estimated number of patients with AD is expected to triple in the US by 2050 to attain 13.7 million people afflicted by the disease, which will bring an increased economic burden to the health care systems (Hebert et al. 2013).

Regardless of the efforts invested in drug development in neurosciences, there remains a high unmet therapeutic need in multiple neurological indications. Drug discovery has been characterized with a high attrition rate, in general, and this is exemplified in CNS, as success rates drop from an average 11–8% (Kola and Landis 2004). Multiple factors contribute to this high attrition and failure rate: lack of adequate animal models, poor crossing of drug through the BBB and insufficient drug penetration into the brain, poor knowledge of pathophysiological mechanisms, increased placebo responses, and, most prominently, lack of specific biomarkers to measure therapeutic effect (Pangalos et al. 2007). However, significant recent progress has been achieved in the biomarker field with the development of the "omics" technologies: genomics, proteomics, metabolomics, lipidomics, and immunological and biological epigenetics (Sethi and Brietzke 2016). The advancement of the neuroimaging techniques has further dramatically contributed to successful drug development in indications such as multiple sclerosis (MS), with 16 disease modifying therapies (DMTs) approved to date. This is considered as quite an accomplishment in view of low success rates of investigated DMTs in more prevalent diseases such as AD and PD. This has led to the increased interest in developing highly targeted biomarkers which will both contribute to reducing attrition and costs as well as, potentially, speed up the drug development processes (Frank and Hargreaves 2003).

This chapter reviews the more widely assessed biomarkers in the major neurological diseases including AD, PD, MS, and neuro-orphan indications.

Biomarker Definition

As per the Biomarkers Definitions Working Group, a biomarker is a characteristic that is objectively measured and evaluated as an indicator of normal biologic processes, pathologic processes, or biological responses to a therapeutic intervention. Change in biomarkers after treatment will help identifying a treatment benefit and safety issues potentially related to that same treatment. An exemplary biomarker should be accessible, reproducible, quantifiable, and easy to apply in the studied patient populations. A marker can be a surrogate to replace a clinical endpoint and give indication on disease modification versus symptomatic effect. A PD marker is a biomarker of pharmacologic response. Both surrogate and PD markers are a subset of biomarkers (Biomarkers Definitions Working Group 2001; FDA 2014).

Biomarkers in Neurological Disorders

The need for the development of DMTs in multiple neurological diseases warrants the use of targeted biomarkers that are both diagnostic of early disease and predictive of drug effect. The inclusion in AD clinical trials of prodromal or preclinical/asymptomatic patients necessitates the incorporation of multiple biomarkers both to permit identification of such subjects and to enhance the chances of proving a subsequent drug effect. A combination of biochemical, neuroimaging, and clinical assessments has become a standard approach in clinical trials, especially in early stages of drug development. Choosing the most optimal combination of biomarkers is critical in early proof-of-concept studies to increase clinical trial success. The selection of the most appropriate biomarkers, whether in isolation or combination, becomes even more relevant when

testing DMTs to differentiate symptomatic effects when compared with longer-term disease modification effects.

General Biomarkers in Neurology

This section summarizes the frequently used biomarkers in neurology while focusing more specifically on the major diseases such as AD, PD, and MS (Giacomelli et al. 2017; Henley et al. 2005; Silva and Furie 2009).

Cerebro-Spinal Fluid

Although lumbar punctures are invasive procedures and a number of findings remain to be confirmed as reliable, cerebral-spinal fluid (CSF) biomarkers are relevant in indications such as AD where a decrease in Abeta 1-42, as well as an increase in Tau is observed (Ittner and Götz 2011). In addition, a reduction in total alpha-synuclein levels in PD, the increase of neurofilaments levels in amyotrophic lateral sclerosis (ALS), as well as the increase in various inflammatory markers in MS have been explored in many studies. The importance of these biomarkers and the need for their use to monitor drug effect warrants the integration of this procedure in clinical trials, while mitigating the invasive aspect of lumbar punctures (LPs).

In AD, for those sites who do not have access to PET scanning facilities and/or access to the PET ligands for amyloid detection, one option used by some pharmaceutical companies is to use CSF for confirmation of amyloid levels as an entry criterion for clinical trials that are utilizing an amyloid-targeting investigational product (ClinicalTrials.gov). As mentioned below, compliance is increasing with this modality and is generally higher in subjects with preclinical and/or prodromal AD than in subjects with mild to moderate disease (IQVIA proprietary database). LPs are not usually standard of care for AD subjects for most countries, and as such, it has taken a number of years to ensure compliance with this assessment (Hughes et al. 2014). However, in some countries, e.g., Sweden, LPs are routinely performed in the majority of subjects presenting with AD or possible AD. Recently, with the advent of the potential DMTs in PD, LPs are now being utilized in these early stage trials, leading to greater challenges than those facing AD trials more than 10 years ago. As CSF collection is not a standard of care in the PD population, other noninvasive procedures are being studied to avoid multiple LPs in these patients.

Neuroimaging

Multiple imaging techniques have contributed significantly to the advancement of drug development in Neurology. The demonstration of magnetic resonance imaging (MRI) as a highly reliable biomarker is most prominent in MS, allowing the approval of multiple drugs, which is without precedent in Neurosciences. The most frequently used MRI sequences are listed here:

MRI with gadolinium: T1 sequences with or without gadolinium enhancement are essential in monitoring the anti-inflammatory drug effect in MS, as gadolinium enhancement is a direct measure of the BBB disruption due to the inflammatory processes.

Structural and Volumetric MRI: Increasingly used to assess the changes of specific anatomical structures of the brain as a result of pathological disease processes. The measuring of medial temporal and hippocampal atrophy in AD is a reliable marker of disease progression (Riascher et al. 2009).

Other MRI techniques such as functional MRI (fMRI), magnetic resonance spectroscopy (MRS), and diffusion-weighted magnetic resonance imaging have been studied in AD and mild cognitive impairment (MCI), Huntington's disease (HD), PD, MS, and stroke.

Positron emission tomography (PET): PET scanning is widely used in a number of neurological indications including AD and MCI (reduced glucose uptake), PD, and HD. FDG PET has been shown to have a good sensitivity to detect brain dysfunction and early changes in AD and to follow its evolution over time (de Leon et al. 2001).

SPECT/DaTScan: This imaging technique uses a transporter to bind to the DAT in the striatum and then SPECT visualizes the amount of transporter present. It allows for the differential diagnosis between PD, essential tremor, multiple system atrophy (MSA), and other Parkinsonian syndromes.

Genomics

The advancement in genetic research has been instrumental in the understanding of multiple neuro-orphan diseases such as ALS (SOD1, C9orf), spinal muscular atrophy (SMA: SMN1 gene), and Duchenne muscular dystrophy (DMD: various exons). This has allowed the development of genome-targeted therapies, with recent significant therapeutic successes in SMA. Further research is critical to bring therapies to patients with rare disease.

Peripheral Biomarkers

Multiple biomarkers have been explored in peripheral fluid such as plasma, red blood cells, and saliva. Confirmation of their reliability is needed to avoid invasive CSF collection in patients in whom LPs are not part of the standard of care. An example of the relevance of peripheral biomarkers is Parkinson's disease as described next.

Clinical Biomarkers

The change in clinical biomarkers to monitor drug effect is a required primary objective of many pivotal trials. The mainstay of assessment for clinical trials in AD is still the use of a number of neurocognitive batteries, with changes in other biomarkers being used as secondary or exploratory endpoints. The use of the Expanded Disability Status Scale (EDSS) remains the gold standard in the observance of disease disability in MS as a hallmark of disease progression. In Parkinson's disease, multiple scales and corresponding subscales, of which most prominently the UPDRS, have been used to assess the effect of drugs in development, on motor function, and for drug-induced complications. Functional rating scales have also been widely used in a variety of neurological clinical trials, as a primary or secondary endpoint.

Electrophysiology

Electroencephalography (EEG), a noninvasive and cost-effective procedure, has proven to be a reliable biomarker in diseases such as AD, epilepsy, pain, PD (REM behavior disorder), and autism. The progress in EEG technologies will provide superior evaluation of central drug effect (Jobert et al. 2012). Although not widely performed in pivotal studies, its use is cost-effective and noninvasive.

Electromyography and electrical impedance myography (Rutkove et al. 2012), the motor-unit number estimation (MUNE), allows the monitoring of disease progression in ALS (Bowser et al. 2006).

Biomarkers in Alzheimer's Disease

There have not been any successful new drugs developed for AD for more than a decade. From 2002 to 2013, 96.4% of the 244 agents assessed in more than 1,000 clinical trials in AD subjects failed to achieve their primary endpoints (Ousset et al. 2001). These trials have been increasingly complex, requiring increased number of sites and assessments and a lower number of subjects overall per site, and subsequently have resulted in higher screen failure rates and, oftentimes, drop-out rates (Ousset et al. 2001). In addition, these trials are becoming more costly, and, in particular, the cost for screen failure rates is prohibitive for many companies.

The combination of biomarkers has been used not only for secondary or exploratory endpoints but also to enrich the clinical trial population, allowing for the most appropriate subjects to be exposed to the clinical trial investigational products and thus maximizing the probability of seeing a signal in the trial data.

The use of biomarkers as entry criteria increased following the data released in 2010 on Semagacestat (Doody et al. 2013) and on Bapineuzamab (Salloway et al. 2014). Both investigational products targeted amyloid, and the

ultimate data analysis revealed that approximately 30% of the recruited suspected mild-moderate AD subjects did not, in fact, have amyloid "confirmed" AD. This enrichment technique has proven to be necessary in large pivotal AD programs and has been found to be useful in successfully identifying AD patients who have a higher chance to benefit from an amyloid-targeting agent.

Subsequently, trials that had an investigational product targeting the amyloid pathway required that subjects have a baseline amyloid positivity before trial entry to ensure that the investigational product did have a target for engagement. However, for the mild to moderate population, this added a further 30% onto the screen failure rate of AD trials, in addition to the 20–30% already observed (Hughes et al. 2014). This effect of enrichment is even more pronounced in the earlier stages of AD with prodromal AD having, overall, a screen failure rate of 70–80%, and preclinical AD being even higher at 90% or more (Hughes et al. 2014). This all leads to an overall effect of decreasing recruitment rates at sites and increasing trial duration and costs.

Amyloid is assessed either via PET imaging with a fluorine-18 labeled ligand or via a lumbar puncture and collection and analysis of CSF. There are basically 3 amyloid ligands available: Avid (Florbetapir), GE (Flutametamol), and Piramel (Florbetaben). The geographical location of labeling centers dictates the site and country distribution for each imaging modality. Thus, there are many global sites and countries where the AD subjects are unable to participate in trials which utilize amyloid PET imaging as a screening tool as these countries either do not have access to a PET scanner and/or a cyclotron. The ligand is generally provided to the sites ready-labeled for immediate administration to the subject and the time from labeling, QC, and subject administration is approximately 4 h due to the half-life of the fluorine-18 (Jobert et al. 2012).

Alternatively, amyloid positivity can be accessed via LPs, which brings with it its own challenges. A review by IQVIA in 2013 of compliance with at least 2 LPs in 12 AD programs, which recruited in excess of 7500 subjects, showed a global compliance of 8% in 2007 increasing to 50% by 2012 (IQVIA proprietary database). This compliance rate is steadily increasing, but is still not ideal (Jobert et al. 2012).

These imaging and CSF biomarkers are being used primarily to ensure that the appropriate subjects are being recruited into the trials. They are not being used as primary endpoints as the regulators still request that a clinical trial in AD shows a positive clinical benefit to a subject and not just a change in nonclinical biomarkers.

An ongoing academic trial, however, utilizes a combination of biomarkers as primary endpoints. This trial is enrolling study participants who are biological adult children of a parent with a mutated gene known to cause dominantly inherited AD. Such individuals may or may not carry the gene themselves and may or may not have disease symptoms. However, it is known that the deposition of amyloid builds up some 10–20 years before clinical symptoms are manifest. Therefore, while utilizing potential disease modifying drugs which remove amyloid or prevent its deposition or build up is being used as surrogate endpoint as for many subjects, the neurocognitive decline is minimal at this very early stage (Bateman et al. 2017). This trial, which is assessing the impact of three investigational products versus placebo, is still recruiting and is being seen almost as a proof-of-concept trial to assess the utility of various AD biomarkers as a key outcome.

Other biomarkers used in AD trials include the assessment of Tau and p-Tau levels in the CSF, and recently, a number of Tau PET ligands have been developed and are now in clinical trials. Tau pathology appears to closely correlate with neurodegeneration and onset of clinical dementia, with dynamic changes in tau pathology being evident at the minimal cognitive impairment (MCI) stage (Lambracht-Washington and Rosenberg 2013). Therefore, this represents an interesting target for pharmaceutical companies, and there are a number of products in early stage clinical trials looking at the possibility of actively or passively immunizing against the various tau biomarkers (IQVIA proprietary database).

Recent advances in AD biomarkers include a race to develop a blood test for amyloid, as PET imaging with an amyloid ligand is not practical on a global scale. If confirmed as a reliable biomarker, this will provide a simple, cost-effective assessment to be used for amyloid level assessment for detection of patients with either prodromal or very early disease and thus enable a physician to specifically target therapy with an appropriate drug.

Biomarkers in Parkinson's Disease

Parkinson's disease (PD) is the second most prevalent neurodegenerative disorder after Alzheimer's disease and affects approximately 1% of the population older than 60 (Tysnes and Storstein 2017). The main hallmark of the disease is loss of dopaminergic neurons in the mesencephalon resulting in the clinical presentation of the disease characterized by 3 cardinal symptoms: bradykinesia, extrapyramidal rigidity, and resting tremor. By the time the patient becomes symptomatic, over 60% of dopaminergic neurons would have been lost, hence, the importance of implementing disease modifying therapy strategies very early in the disease.

The treatment of PD remains symptomatic and suboptimal as a result of treatment complications. Developing disease-modifying therapies that would slow disease progression outside of isolated symptomatic relief is essential. For this effect, reliable biomarkers allowing early disease detection are fundamental. The ideal biomarker in PD would aim at early detection of disease at prodromal stage, confirm differential diagnosis with other diseases such as PSP or MSA, and allow monitoring of disease progression (Shapira 2013).

There have been important advances in biomarkers in PD in recent years. The Parkinson's Progression Markers Initiative (PPMI), a multicenter, observational study, is currently underway, and its main objective is identifying imaging, biological, clinical, and behavioral assessments biomarkers of disease progression in a standardized way (Parkinson Progression Marker Initiative 2011).

Multiple imaging techniques have found to be relevant biomarkers in PD, of which the most prominent is dopamine transporter (DAT) imaging. It allows the identification of significant reduction of dopaminergic neurons. The usefulness of this technique for disease progression remains however to be further confirmed (Wang et al. 2013). Alpha-synuclein, a protein found in Lewy bodies, the pathological hallmark of PD, has emerged as a very relevant biomarker. Levels of total alpha-synuclein have been found to be decreased in CSF, while oligomer forms of this protein have been reported to be increased. Recent findings seem to indicate that measuring the levels of alpha-synuclein might not only be diagnostic of PD but serve as a marker for disease progression (Wang et al. 2013). The assessment of peripheral alpha-synculein blood and Saliva levels could be utilized as biomarkers, avoiding invasive procedures (Devic et al. 2011). Significant advances have been made in the genetic understanding of familial forms of PD (mutations in LRKK2 and PINK genes) that might accelerate the development of effective and targeted therapies (Li and Yang 2011).

Biomarkers in Multiple Sclerosis

The discovery of MRI technology has contributed significantly to the accelerated drug development in multiple sclerosis (MS). Specific sequences such as T1 with and without gadolinium, T2, and FLAIR allow the detection of the severity of inflammation, demyelination, and axonal loss. Following the changes of these parameters provides information demonstrating drug effect on disease activity. Furthermore, whole brain and spinal volumetric measurement identifies the severity of atrophy in specific regions of the central nervous system and hence can be used as a surrogate marker of disease progression (Nair et al. 2013).

CSF, although an invasive procedure, is valuable for marker collection of inflammation (Oligoclonal bands), demyelination (Myelin Basic

Protein, MBP), and axonal loss (Neurofilaments) (Giovannoni 2006). The levels of JCV in CSF are a safety marker to identify subjects at potential risk of developing progressive multifocal leucoencephalopathy (PML) while on potent immunomodulators/immunosupressors.

Optic coherence tomography (OCT) measures retinal nerve fiber layer (RNFL) thickness and macular volume. This technique can be used as a marker of neuroprotective effect of certain drugs (Villoslada 2010).

The Expanded Disability Status Score (EDSS), the Multiple Sclerosis Functional Composite (MSFC) that includes the Paced Auditory Serial Addition Test (PASAT), and the Symbol Digit Modalities Test (SDMT) are recognized clinical biomarkers of both physical and cognitive disability in MS.

Biomarkers in Neuro-Orphan Indications

Biomarkers in Amyotrophic Lateral Sclerosis (ALS)

Multiple advances have been made in the past years in identifying biomarkers for amyotrophic lateral sclerosis (ALS). Significant progress has been observed in the genomic, neuroimaging (MRI, Diffusion Tensor Imaging, PET), and electrophysiologic marker field. Motor unit number estimation (MUNE), electrical impedance myography, and the neurophysiological index are the most prominent electrophysiological techniques that can be useful in disease progression monitoring (Bowser et al. 2011).

Recent findings have revealed increased neurofilament (NFL) levels in the CSF of ALS patients compared to controls as a marker of neurodegeneration, increased blood levels of creatinine, as well as NFL, and NOGO-A strong expression in muscles (Chen and Shang 2015).

Further validation of the different biomarkers and their utility, in isolation or in combination, in disease diagnosis and progression, is needed.

Biomarkers in Huntington's Disease

Huntington's disease is an autosomal dominant neurodegenerative condition caused by a CAG trinucleotide expansion in the Huntingtin gene (HTT) (The Huntington's Disease Collaborative Research Group 1993). A new wave of investigational products is currently under investigation in this disease arena, which is aimed at core disease mechanisms (Chen and Shang 2015). The development of new therapies depends on a robust and in depth understanding of the pathogenesis of this disease and the ability to translate this knowledge into pharmacodynamics biomarkers. Currently there is much interest in the quantification of mutant Huntingtin (mHTT) in the CSF and PET imaging targeting Huntingtin (HTT) as a potential biomarker (Mestra and Sampaio 2017). Although the actual goal of DMT is to lower the levels of the HTT protein, the molecular targets of the intervention are different products of the HTT gene – mostly RNAs or the gene itself. Ongoing trials in this arena include the assessment of various antisense oligonucleotides and RNA interference strategy agents (IQVIA proprietary database).

Summary

Significant progress has been made in the field of biomarkers for neurological disorders. These span from imaging, genetics, molecular, biochemical, clinical to electrophysiological methods. The use of biomarkers in isolation or combination, for diagnostic and predictive purposes in the clinic, while allowing the monitoring of drug effect in a clinical trial setting, has evolved rapidly in recent years. Greater investment and research in this field is still needed to allow for accelerated discovery of therapeutic interventions, in an area with important unmet medical needs.

References and Further Reading

Alavijeh MS, Chishty M, Qaiser MZ et al (2005) Drug metabolism and pharmacokinetics, the blood-brain barrier, and central nervous system drug discovery. NeuroRx 2:554–571

Bateman RJ, Benzinger TL, Berry S, DIAN-TU Pharma Consortium for the Dominantly Inherited Alzheimer Network et al (2017) The DIAN-TU next generation Alzheimer's prevention trial: adaptive design and disease progression model. Alzheimers Dement 13:8–19

Biomarkers Definitions Working Group (2001) Biomarkers and surrogate endpoints: preferred definitions and conceptual framework. Clin Pharmacol Ther 69:89–95

Bowser R, Cudkowicz M, Kaddurah-Daouk R (2006) Biomarkers for amyotrophic lateral sclerosis. Expert Rev Mol Diagn 6:387–398

Bowser R, Turner M, Shefner J (2011) Biomarkers in amyotrophic lateral sclerosis: opportunities and limitations. Nat Rev Neurol 7:631–638

Chen X, Shang F (2015) New developments and future opportunities in biomarkers for amyotrophic lateral sclerosis. Transl Neurodegener 4:17

Calcagno A, Di Perri G, Bonora S (2014) Pharmacokinetics and pharmacodynamics of antiretrovirals in the central nervous system. Clin Pharmacokinet 53:891–906

ClinicalTrials.gov. https://clinicaltrials.gov/. Accessed 19 Sept 2017

Danhof M, de Jongh J, De Lange EC et al (2007) Mechanism-based pharmacokinetic-pharmacodynamic modeling: biophase distribution, receptor theory, and dynamical systems analysis. Annu Rev Pharmacol Toxicol 47:357–400

de Lange E (2013) The mastermind approach to CNS drug therapy: translational prediction of human brain distribution, target site kinetics, and therapeutic effects. Fluids Barriers CNS 10:12

de Leon MJ, Convit A, Wolf OT et al (2001) Prediction of cognitive decline in normal elderly subjects with 2-[(18)F]fluoro-2-deoxy-D-glucose/positron-emission tomography (FDG/PET). Proc Natl Acad Sci U S A 98:10966–10971

Devic I, Hwang H, Edgar JS et al (2011) Salivary alpha-synuclein and DJ-1: potential biomarkers for Parkinson's disease. Brain 134:e178

Di L, Kerns EH (eds) (2015) Blood-brain barrier in drug discovery: optimizing brain exposure of CNS drugs and minimizing brain side effect for peripheral drugs. Wiley, Malden

Dingemanse J, Danhof M, Breimer DD (1998) Pharmacokinetic-pharmacodynamic modeling of CNS drug effects: an overview. Pharmacol Ther 38:1–52

Donovan MD, Boylan GB, Murray DM et al (2015) Treating disorders of the neonatal central nervous system: pharmacokinetic and pharmacodynamic considerations with a focus on antiepileptics. Br J Clin Pharmacol 8:62–77

Doody RS, Raman R, Farlow M, Alzheimer's Disease Cooperative Study Steering Committee, Siemers E, Sethuraman G, Mohs R, Semagacestat Study Group (2013) A phase 3 trial of semagacestat for treatment of Alzheimer's disease. N Engl J Med 369:341–350

Durham TB, Blanco MJ (2015) Target engagement in lead generation. Bioorg Med Chem Lett 25:998–1008

FDA (2014) Guidance for industry and FDA staff qualification process for drug development tools. U.-S. Department of Health and Human Services Food and Drug Administration Center for Drug Evaluation and Research. Silver Spring, MD, USA

Frank R, Hargreaves R (2003) Clinical biomarkers in drug discovery and development. Nat Rev Drug Discov 2:566–580

Giacomelli C, Daniele S, Martini C (2017) Potential biomarkers and novel pharmacological targets in protein aggregation-related neurodegenerative diseases. Biochem Pharmacol 131:1–15

Giovannoni G (2006) Multiple sclerosis cerebrospinal fluid biomarkers. Dis Markers 22:187–196

Hebert L, Weuve J, Scherr P et al (2013) Alzheimer disease in the United States (2010–2050) estimated using the 2010 census. Neurology 80:1778–1783

Henley S, Bates G, Tabrizi S (2005) Biomarkers for neurodegenerative diseases. Curr Opin Neurol 18:698–705

Hughes L, Hayduk R, Vanbelle C (2014) Integrating biomarkers in Alzheimer's disease trials: review of compliance with biomarker assessments in Alzheimer's disease clinical trials. Available from the author upon request

ICH (2017). Pediatric guideline. Available at: http://www.ich.org/products/guidelines/efficacy/efficacy-single/article/addendumclinical-investigation-of-medicinal-products-in-the-pediatric-population.html (accessed 30 November 2017)

Ittner LM, Götz J (2011) Amyloid-β and tau – a toxic *pas de deux* in Alzheimer's disease. Nat Rev Neurosci 12:67–72

Jobert M, Wilson F, Ruigt G, The IPEG Pharmaco-EEG Guidelines Committee et al (2012) Guidelines for the recording and evaluation of pharmaco-EEG data in man: the international Pharmaco-EEG society (IPEG). Neuropsychobiology 66:201–220

Johnson KA, Minoshima S, Bohnen NI et al (2013) Update on appropriate use criteria for amyloid PET imaging: dementia experts, mild cognitive impairment, and education. J Nucl Med 54:1011–1013

Kola I, Landis J (2004) Can the pharmaceutical industry reduce attrition rates? Nat Rev Drug Discov 3:711–716

Lambracht-Washington D, Rosenberg RN (2013) Anti-amyloid beta to tau-based immunization: developments in immunotherapy for Alzheimer's disease. Immunotargets Ther 2:105–114

Li T, Yang D, Sushchky S et al (2011) Models for LRRK2-linked Parkinsonism. Parkinsons Dis 2011:942412

Maharaj AR, Barrett JS, Edginton AN (2013) A workflow example of PBPK modeling to support pediatric research and development: case study with lorazepam. AAPS J 15:455–464

Melham M (2013) Translation of central nervous system occupancy from animal models: application of pharmacokinetic/pharmacodynamic modeling. J Pharmacol Exp Ther 347:2–6

Mestra TA, Sampaio C (2017) Huntington's disease: linking pathogenesis to the development of experimental therapeutics. Curr Neurol Neurosci Rep 17:18

Nair G, Shea C, Crainiceanu C et al (2013) Quantification of multiple-sclerosis-related brain atrophy in two heterogeneous MRI datasets using mixed-effects modeling. Neuroimage Clin 3:171–179

Nielsen JC, Tolbert D, Patel M et al (2014) Vigabatrin pediatric dosing information for refractory complex partial seizures: results from a population dose–response analysis. Epilepsia 55:e134–e138

Ousset P-J, Cummings J, Delrieu J et al (2001) Is Alzheimer's disease drug development broken? What must be improved. JPAD 1(1), 2014 Proc Natl Acad Sci USA, pp 1–7

Pangalos M, Schechter L, Hurko O (2007) Drug development for CNS disorders: strategies for balancing risk and reducing attrition. Nat Rev Drug Discov 6:521–532

Parkinson Progression Marker Initiative (2011) The Parkinson progression marker initiative (PPMI). Prog Neurobiol 95:629–635

Pellock JM, Carman WH, Thyagarajan V et al (2012) Efficacy of antiepileptic drugs in adults predicts efficacy in children: a systematic review. Neurology 79:1482–1489

Pellock JM, Arzimanoglou A, D'Cruz, Pediatric Epilepsy Consortium for Extrapolation et al (2017) Extrapolating evidence of antiepileptic drug efficacy in adults to children $\geq$2 years of age with focal seizures: the case for disease similarity. Epilepsia 58:1686

Politis M, Piccini P (2012) Positron emission tomography imaging in neurological disorders. J Neurol 259:1769–1780

Quintiles proprietary database

Reichel A (2009) Addressing central nervous system (CNS) penetration in drug discovery: basics and implications of the evolving new concept. Chem Biodivers 6:2030–2049

Reichel A (2015) Pharmacokinetics of CNS penetration. In: Di L, Kerns EH (eds) Blood-brain barrier in drug discovery: optimizing brain exposure of CNS drugs and minimizing brain side effect for peripheral drugs. Wiley, Malden, pp 7–36

Riascher SL, Saykin AJ, West JP et al (2009) Baseline MRI predictors of conversion from MCI to probable AD in the ADNI cohort. Curr Alzheimer Res 6:347–361

Rizk ML, Zou L, Savic RM et al (2017) Importance of drug pharmacokinetics at the site of action. Clin Transl Sci 10:133–142

Rutkove S, Caress J, Cartwright M et al (2012) Electrical impedance myography as a biomarker to assess ALS progression. Amyotroph Lateral Scler 13:439–445

Salloway S, Sperling R, Fox NC, Bapineuzumab 301 and 302 Clinical Trial Investigators et al (2014) Two phase 3 trials of bapineuzumab in mild-to-moderate Alzheimer's disease. N Engl J Med 370:322–333

Seifert KD, Wiener JI (2013) The impact of DaTscan on the diagnosis and management of movement disorders: a retrospective study. Am J Neurodegener Dis 2:29–34

Sethi S, Brietzke E (2016) Omics-based biomarkers: application of metabolomics in neuropsychiatric disorders. Int J Neuropsychopharmacol 19:pyv096

Shapira A (2013) Recent developments in biomarkers in Parkinson disease. Curr Opin Neurol 26:395–400

Silva G, Furie K (2009) Biomarkers in neurology. In: Woodbury-Harris KM, Coull BM (eds) Clinical trials in the neurosciences. Karger, Basel, pp 55–61

The Huntington's Disease Collaborative Research Group (1993) A novel gene containing a trinucleotide repeat that is expanded and unstable on Huntington's disease chromosomes. The Huntington's Disease Collaborative Research Group. Cell 72:971–983

Tuntland H, Espehaug B, Forland O et al (2014) Reablement in community-dwelling adults: study protocol for a randomised controlled trial. BMC Geriatr 14:139

Tysnes O, Storstein A (2017) The epidemiology of Parkinson's disease. J Neural Transm (Vienna) 124:901–905

Villoslada P (2010) Focus on biomarkers: biomarkers for multiple sclerosis. Drug News Perspect 23:585–595

Wang J, Hoekstra J, Zuo C et al (2013) Biomarkers of Parkinson's disease: current status and future. Drug Discov Today 18:155–162

Wiltshire HR, Kilpatrick GJ, Tilbrook GS (2012) A placebo and midazolam-controlled phase I single ascending dose study evaluating the safety, pharmacokinetics, and pharmacodynamics of remimazolam. Anesth Analg 115:284–296

Yamamoto S, Karashima M, Arai Y et al (2017) Prediction of human pharmacokinetic profile after transdermal drug application using excised human skin. J Pharm Sci 106:2787–2794