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Pharmacokinetic and Drug Metabolism Properties of Novel Therapeutic Modalities

Brooke M. Rock[1] and Robert S. Foti[2]

[1]Pharmacokinetics and Drug Metabolism, Amgen Research, 1120 Veterans Blvd, S. San Francisco, 94080

[2]Pharmacokinetics and Drug Metabolism, Amgen Research, 360 Binney St, Cambridge, MA 02142

Running Title Page: PK Properties of Novel Therapeutic Modalities

Corresponding Authors:

Brooke M. Rock, Ph.D.
Principal Scientist
Pharmacokinetics and Drug Metabolism
Amgen, Inc.
1120 Veterans Boulevard
South San Francisco, CA 94080
brooke@amgen.com

Robert S. Foti, Ph.D.
Principal Scientist
Pharmacokinetics and Drug Metabolism
Amgen, Inc.
360 Binney Street
Cambridge, MA 02142
rfoti@amgen.com

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Abbreviations: ADC, antibody-drug conjugate; PK-PD, pharmacokinetic-pharmacodynamic; ON, oligonucleotide; siRNA, short interfering RNA molecules; ASO, antisense oligonucleotide; SM small molecule; mAb, monoclonal antibody.

Abstract

The discovery and development of novel pharmaceutical therapies is rapidly transitioning from a small molecule-dominated focus to a more balanced portfolio consisting of small molecules, monoclonal antibodies, engineered proteins (modified endogenous proteins, bispecific antibodies and fusion proteins), oligonucleotides and gene-based therapies. This commentary, and the special issue as a whole, will aim to highlight these emerging modalities and the efforts underway to better understand their unique pharmacokinetic and ADME properties. The manuscripts highlighted herein can be broadly grouped into those focusing on the ADME properties of novel therapeutics, those exploring targeted-delivery strategies and finally, those discussing oligonucleotide therapies. It is also evident that while the field in general continues to progress towards new and more complex molecules, a significant amount of effort is still being placed on antibody-drug conjugates (ADCs). As therapeutic molecules become increasingly complex, a parallel demand for advancements in experimental and analytical tools will become increasingly evident, both to increase the speed and efficiency of identifying safe and efficacious molecules while simultaneously decreasing our dependence on in vivo studies in preclinical species. The research and commentary included in this special issue will provide researchers, clinicians and the patients we serve more options in the ongoing fight against grievous illnesses and unmet medical needs.

Significance Statement

Recent trends in drug discovery and development suggest a shift away from a small molecule-dominated approach to a more balanced portfolio that includes small molecules, monoclonal antibodies, engineered proteins and gene therapies. The research presented in this special issue of Drug Metabolism and Disposition will serve to highlight advancements in the understanding of the mechanisms which govern the pharmacokinetic and drug metabolism properties of the novel therapeutic modalities.

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Introduction

The speed at which science is evolving has unveiled new biological pathways and novel drug targets (Santos et al., 2017). In many cases, modulating new targets can require exploring pharmacology beyond classic small molecule-target interactions, leading to an influx of new therapeutic modalities. A look at the new drugs approved by the U.S. Food and Drug Administration since 1980 illustrates a rapidly emerging change from approvals dominated by small molecules (prior to 2000) to a more balanced portfolio of small molecules and "non-small molecule" drugs such as monoclonal antibodies, engineered proteins, oligonucleotides and vaccines (Figure 1a) (Reichert, 2003; Mullard, 2018; Morrison, 2019). Similarly, in the last ten years, the list of top selling drugs in North America has swung from being dominated by small molecules to now listing 8 of the 10 top selling drugs in the United States as protein therapeutics (Quan et al., 2018; Wineinger et al., 2019). Indeed, an analysis of the total sales of the top ten drugs in each year from 2010 to present shows a decreasing dependence on small molecule revenue and a rapid shift towards sales of monoclonal antibodies and engineered proteins (Figure 1b) (Bartholow, 2015; Drugs.com, 2019; Philippidis, 2019).

As molecular biology and genetic techniques continue to evolve, modalities beyond typical small and large molecules are rapidly finding their way into the drug discovery pipeline. This evolution is requiring drug metabolism and pharmacokinetic researchers to develop new tools to understand the absorption, disposition, metabolism and excretion of these new chemical entities, as described in the review by Datta-Mannan included in this issue (Datta-Mannan, 2019). Of particular challenge here is the fact that many of these analytical and experimental tools were developed to characterize monoclonal antibodies, though emerging research suggests that many of the newer, highly-engineered therapeutic proteins may not be subject to the same disposition mechanisms as unmodified antibodies (Vugmeyster et al., 2012; Ueda, 2014). In this issue of *Drug Metabolism and Disposition* a series of primary research articles and review articles are presented to highlight the importance of understanding new modalities from an ADME scientist's perspective. This issue focuses on the following three areas: (1) Biodistribution and

Pharmacokinetic Considerations of New Modalities, (2) Targeted Drug Delivery and (3) Oligonucleotide Therapeutics. In addition, this issue contains a review article with a focus on different mechanisms that influence the disposition of peptides and monoclonal antibodies (Datta-Mannan, 2019).

Biodistribution and Pharmacokinetic Considerations of New Modalities

Establishing a pharmacokinetic-pharmacodynamic (PK-PD) relationship with small and large molecules has traditionally been performed from measuring drug concentration from the blood compartment and linking that concentration to a response (Bueters et al., 2013; Singh et al., 2015; Glassman and Balthasar, 2016). However, new technologies in targeted delivery have required ADME scientists to think beyond the blood compartment for drug and pharmacodynamic measurements to aid in the development of translational models, especially for pharmacological targets expressed outside of the vasculature (Shah, 2015; Glassman and Balthasar, 2016; Tibbitts et al., 2016). In many cases, understanding the biodistribution and exposure to the active form of the molecule at the therapeutic site of action for a new modality is critical to establishing a PK-PD relationship (Lin, 2009; Shah and Betts, 2013; Conner et al., 2014; Glassman et al., 2015). In vitro assays that can provide information on the in vivo pharmacokinetic behavior of a molecule prior to conducting in vivo studies are crucial to inform design of new modalities and can increase the speed of new modalities reaching clinical trials (Xu and Vugmeyster, 2012; Pearson and Rock, 2015). Described in this issue is a novel in vitro assay by Foti et al. which informs how new chemical entities such as peptide-antibody conjugates will behave in vivo and provides a basis upon which to prioritize molecules for in vivo studies (Foti et al., 2019). The advancements of sensitive bioanalytical techniques have also increased the ability to measure drug at the site of action (Foti et al., 2015). To that end, the manuscript by Zhang et al. describes the importance of measuring drug concentration in the correct tissue compartment (e.g. site of action) along with the implications of asymmetric drug distribution (Zhang et al., 2019c). Looked at from a wholistic view point, integrating the results of in vitro and in vivo biodistribution experiments will lead to a stronger translational understanding of new modalities, but will also require advancements to many of our current modelling approaches. The manuscript by Li et al. defines a

mathematical model to understanding PK-PD in a mouse tumor model with different antibody fragments and as such, serves as a good example of a parameterized model which can inform the required amount of drug at the site of action to achieve efficacy (Li et al., 2019).

Targeted Drug Delivery

The aim of a targeted drug delivery approach is to concentrate the active drug at the site of action while reducing exposure to other regions of the body (Iqbal et al., 2017; Srinivasarao and Low, 2017). The hallmark examples of targeted drug delivery are antibody-drug conjugates (ADCs), where a cytotoxin (e.g. small molecule) is covalently attached to a monoclonal antibody targeting the tissue of interest and ultimately increasing the efficacy and therapeutic index of the cytotoxin. There are numerous publications on the ADME properties of ADCs which affect both their biodistribution and efficacy (Bornstein, 2015; Hamblett et al., 2016; Kraynov et al., 2016). For therapies that are designed to directly deliver to the site of action it is also important to understand the blood stability as well as the rate of delivery, as many such molecules utilize linker strategies that may be susceptible to degradation by proteases found in circulation (Tsuchikama and An, 2018). The two included manuscripts by Zhang et al. define different linkages to either peptides or antibodies which enables the effective delivery of the warhead (Zhang et al., 2019a; Zhang et al., 2019b). These authors also point out elements to consider when measuring the drug stability either in vitro or in vivo. Directed drug delivery has also enabled the success of oligonucleotides as therapeutics (Zimmermann et al., 2017) as discussed in the following section.

Oligonucleotide Therapeutics

Oligonucleotide (ON) therapeutics utilize a biological process known as RNA interference (RNAi), in which short interfering RNA molecules (siRNAs) or antisense oligonucleotides (ASOs) neutralize a target

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mRNA sequence in order to prevent subsequent protein translation (Nair et al., 2017; Andersson et al., 2018). While the potential applications of ASOs and siRNAs to the field of therapeutics is vast, understanding the underlying delivery efficiency and relative tissue distribution remains a significant challenge. It is well established in the ON field that sensitive bioanalytical assays are required to study the ADME properties of ONs (Humphreys et al., 2019a; Thayer et al., 2019). Presented herein is an overview by Post et al. of the ADME properties for an ASO from preclinical species to clinical outcomes (Post et al., 2019). In addition to understanding the translation of ON therapeutics, there is limited information on the potential for drug-drug interactions, or the potential effects of protein binding, topics addressed herein by Ramsden et al. and Humphreys et al., respectively (Humphreys et al., 2019b; Ramsden et al., 2019). The therapeutic potential of ON therapeutics is rapidly increasing with the development of new delivery techniques, as evidenced by the multitude of ongoing clinical trials (Titze-de-Almeida et al., 2017; Zimmermann et al., 2017; Saw and Song, 2019). As the field continues to evolve, ADME scientists will have the opportunity to inform the design of newer generation molecules, specifically in regard to the kinetics of drug delivery and impact of co-medications on therapeutic treatments.

Conclusion

In the past decade the expansion of biology and genetics has increased the need for ADME scientists to expand the tools available to underwrite the absorption, disposition, metabolism and excretion of new therapeutic modalities. It is vital to understand and characterize the ADME properties of a new chemical entity that ultimately lead to a safe and efficacious dose (e.g. acceptable therapeutic index). The articles presented herein underscore the importance of integrating in vitro and in vivo experiments in the assessment of novel modalities. Perhaps, one of the greatest challenges in drug discovery is the generation of knowledge that is required to design and develop new modalities and the novel experimental approaches needed to characterize such modalities. ADME scientists across industry and academia are now faced with a unique opportunity and responsibility to continue exploring and discovering new approaches to keep up with and

advance the ever-changing face of therapeutic modalities. This special issue of Drug Metabolism and Disposition on the Pharmacokinetic and Drug Metabolism Properties of Novel Therapeutic Modalities will serve to highlight some of these emerging tools that will be critical in transforming today's experimental molecules into tomorrow's successful therapeutics.

Authorship Contributions

Participated in Research Design:

Conducted Experiments:

Contributed New Reagents or Analytical Tools:

Performed Data Analysis:

Wrote or Contributed to the Writing of the Manuscript: Rock and Foti

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Figure Legends

Figure 1. Recent drug approvals by the U.S. Food and Drug Administration (A) and sales of the top ten best-selling drugs in each year point towards a more balanced therapeutic portfolio (B).

Prior to 2000, U.S. FDA drug approvals consisted of mainly small molecule (SM) therapeutics, though recent trends suggest a more equal distribution between small molecule and biologic approvals (A). Similarly, prior to 2010, the top ten selling drugs in a given year primarily consisted of small molecules, with revenue in recent years being more heavily weighted towards monoclonal antibodies (mAb) and engineered proteins (B).

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Figures.

Figure 1.