BOOK REVIEW

Developability of Biotherapeutics: Computational Approaches. Edited by Sandeep Kumar and Satish K. Singh

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Developability of Biotherapeutics: Computational Approaches aims to lay out the computational tools that are now available to help address the problem of developability for biotherapeutics, in particular antibodies and their derivatives. As aptly described in the foreword by Professor Trout (Massachusetts Institute of Technology), to design a drug it is necessary to do more than design a molecule that binds to a target of interest; it is also vital that the molecule interacts as we wish with other molecules within the body, as well as having suitable properties for manufacture and delivery. The book is written to appeal to those already engaged in drug development, but at a level that enables those who have not considered biotherapeutics before to gain a ready familiarity with the topic. In particular, many of the chapters highlight the differences between small molecules and biotherapeutics and the resulting challenges and opportunities.

The book is divided in 2 main sections. Section I of the book includes 6 chapters covering basic principles and technologies, while section II focuses on practical applications.

Chapter One, by Kumar, Robins, Buck, Hickling, Thangakani, Satish, Singh and Gromiha (Pfizer, University of Madras and Indian Institute of Technology, Madras), discusses the applications of computation in biological drug discovery. This is a wide-ranging chapter touching on several subjects briefly. It starts with an overview of the growing importance of biologics as therapeutics, particularly those based on antibodies. This growth market is placed alongside the increasing desire to make evidence-based decisions in the development of novel biologics, building from and using the information that is now routinely captured in laboratory information management systems. The chapter offers a challenge to what the authors state is the current paradigm for biologics development, where firstly every attempt is made to generate a molecule with high binding affinity toward a target and then only once such a binder has been identified is it screened for developability. The authors pose the question as to whether such separation is a good model for drug discovery in the biologics world, particularly given the size and structural complexities of these molecules. The chapter then goes on to list and discuss many of the potential developability issues of biologics, including aggregation, degradation and adverse immunogenicity. These sections give a preliminary overview, with more detail on many of these issues offered in the following chapters. In their conclusions they reiterate that the “application of computational modeling and simulation during the early stages of biologic drug discovery and formulation can facilitate developability risk assessment for the biopharmaceutical candidates.”

Chapter Two by Dixit (Zymeworks) discusses computational methods in the optimization of biologic modalities. The chapter describes how the increasing knowledge base around the development of biologics coupled with improving computing hardware and software design present real opportunities in our ability to optimize biologics. They discuss the possibility of rational data-driven optimization. The computational technique under focus is the use of molecular simulation to calculate properties of potential biologics. In the early sections of this chapter the innate link between structure and function is given, followed by a brief summary of the functional form of a standard molecular simulation force field. The following sections go through areas in which computation may aid in protein therapeutic optimization, from target discovery in a systems biology approach to modalities of action and pharmacokinetic optimization. There are also short sections on more unusual types of antibodies, such as bispecific and heterodimeric protein domains. The final sections give short introductions to antibody variable domain modeling and the drug delivery system.

In Chapter Three, Agrawal (Amgen) and Chennamsetty (Bristol-Myers Squibb) focus on aggregation and stability of biopharmaceutical drug products. The chapter outlines our current understanding and the computational tools that are available for the prediction of sites that are prone to aggregation, oxidation and deamidation. The majority of the chapter concentrates on aggregation because it is such a key problem and because this is where much computational effort has been devoted to date. The computational studies as described in section 3.1 can be split broadly into 2 intertwined types: 1) those that attempt to understand the mechanism of aggregation and 2) those that attempt to predict aggregation, in particular aggregation prone regions (APRs). Table 3.1 gives a long and very useful list of computational tools for the prediction of APRs in peptides and proteins; the accompanying text describes them in more detail and comments on those that have been used in the prediction of APRs in biotherapeutic
molecules such as monoclonal antibodies (mAbs). This leads naturally into a discussion section concerning designing changes that should mitigate aggregation. Section 3.2 then discusses protein chemical modifications, in particular computational modeling tools that aim to predict potential post-translational modifications and degradation sites based on protein sequence or structure. At the conclusion of the chapter the authors describe how computational tools come at a very low cost and can be predictive of aggregation or degradation sites within a biotherapeutic molecule, harking back to the first chapter in arguing that such tools should be used early in the drug discovery process to help engineer out such sites. Early use of such tools has the potential to reduce costs, as well as shortening the time it takes to bring a biopharmaceutical drug to patients.

In Chapter Four, Jones, Karle and Baker (Antitope and Novartis) consider the topic of preclinical immunogenicity risk assessment. Many biotherapeutics, even those developed from human proteins, elicit anti-drug antibodies (ADAs) in patients. These ADA responses can have a significant impact on the efficacy of a treatment. The first section of the chapter gives an outline of the biology around the immune response, which is brief but contains enough detail to allow the reader to grasp both the complexity of possible problems and some of the areas where solutions might be found. Section 4.2 concentrates on the first of these, T-cell epitope prediction (Table 4.1 gives a summary of freely available web-based tools for the prediction of peptide binding to HLA class II molecules). The authors catalog how such methods work and their limitations, in particular the fact that all tools base predictions on binding a 9-mer within the HLA class II binding groove. This is an important limitation as additional interactions outside the 9-mer are known to have significant influence. Using a case example, the authors show that the tools are not in full agreement and are influenced by the thresholds chosen by users (Fig. 4.2 gives a clear illustration of the problem). Toward the end of the chapter the authors give a short introduction to methods for B-cell epitope prediction discussing both conformational and linear epitopes, but note that removing B-cell epitopes from a protein is very difficult due to the highly adaptive nature of the antibody response.

Chapter Five, by Singh, Tiwari, Abraham and Zutshi (Pfizer and Merck), specifically focuses on mechanistic pharmacokinetics (PK)/pharmacodynamics (PD) models that can be employed when selecting a lead by guiding affinity requirements, as well as in predicting human PK and starting dose for first-in-human (FIH) trials. The models can also help to establish proof of mechanism in humans by projecting pharmacologically active or efficacious dose levels. The opening sections of the chapter discuss natural antibody isotypes, followed by a description of the general absorption, distribution, metabolism, and excretion (ADME) properties of mAbs. Table 5.3 provides a guide to the differences between biologics and small molecules in terms of their ADME properties, useful for those with backgrounds in small molecule drug development. There is a brief discussion of systems pharmacology models followed by a more detailed one on current mathematical modeling techniques for the calculation of PK/PD properties of mAbs. The authors then discuss the prediction of human PK and the starting dose projections for FIH trials. The authors conclude by highlighting the lack of data currently available, in particular quantitative data, about the interaction networks in the human body. They argue that, as such data increases in availability, PK/PD models will move from empirical to more mechanistic.

In Chapter Six, Chaudhri (Lawrence Berkeley National Lab) describes how computational methods might help with the challenges in high concentration biopharmaceutical drug delivery. The chapter opens with an explanation for the need for high doses and the problems this causes. The author indicates how the complications of aggregation and self-association of proteins in high concentration can be explored by the use of molecular simulations. In this chapter, the use of both coarse-grained and atomic level procedures is discussed. The results from coarse-grained work have led to the idea that the unusually high viscosities of mAbs in high concentration is caused by short-range electrostatic interactions. Computational models that can be quantitatively compared to viscosity measurements can only be achieved if computational models incorporate explicit solvent and higher order electrostatic effects. The chapter lists several of the challenges still facing accurate calculation of electrostatic interactions of high concentration protein solutions.

Overall, the first 6 chapters of the book are a useful starting compendium of the computational and molecular modeling techniques that are now available and are able to make real contributions toward biopharmaceutical developability.

Section II presents 4 perspectives from leaders in biopharmaceutical development espousing on how their respective organizations approach developability as they move biotherapeutics molecules from discovery to the clinic. Perhaps not surprisingly, there is substantial overlap among the 4 perspectives, with all pointing out the importance of key properties like expression levels (titers), physical and chemical stability, and solubility. None-the-less, each chapter does include unique insights or approaches, and these aspects will be emphasized below.

Chapter Seven by Hartmann and Kocher (Novartis) describes a hierarchical process of assessment mapped to an intuitive “traffic light” scoring of green, yellow and red lights, to signify how different molecules advance along the development pathway. From the start, the authors emphasize the notion of “fit” into available platform approaches. Some “difficult” molecules may be only so in the context of a platform, but nevertheless, all factors being equal, it behoves one to de-prioritize such entities within a larger portfolio perspective. As the authors indicate, mAbs enter the assessment phase from multiple discovery approaches that include animal immunization, phage display, and immortalization of antibody-producing cells. The triage includes an early selection phase where, typically, around 50 molecules are evaluated. Critical risk factors here include expression and aggregation. Poor expression titers, usually in transient HEK transfection, may not be highly predictive of behavior in Chinese hamster ovary (CHO) cells that are stably transfected. However, with many options to choose from and in the interest of timelines, molecules showing such poor expression may be de-prioritized earlier on. Other factors, denoted “cumulative,” are not individually critical or leading to go-no-go decisions, but as a composite may ultimately determine which molecules move to the next stage. These factors include isoelectric point, thermal stability, behavior in hydrophobic...
interaction chromatography (HIC), Fd: LC interface stability, among others. The more comprehensive, final, profiling phase is usually applied to a handful of molecules and it aims to select a lead and a backup. Here productivity (titer) in CHO stable pools is assessed, together with deeper molecular profiling, assessment of thermal stability by more refined techniques (differential scanning calorimetry vs. Thermofluor in early evaluation), pre-formulation scanning and in vivo fitness. A schematic example of 4 such leads is presented with a detailed discussion of the thought process that landed on a lead and backup choice.

In Chapter Eight, Angell, Ketchem, Daris, O’Neill and Gunasekaran (Amgen) outline their approach to developability assessment. There is a prominent place for engineering, particularly using the rational approach of generating and evaluating individual variants around a lead (vs. library approaches). Explicit computational modeling of 3D structures of the Fv of antibodies of interest is carried out. The resulting models, usually an ensemble for a given individual input sequence, are used to help assess, and when possible “engineer-out,” undesirable traits like deamidation and isomerization sites. It is acknowledged that the presence of certain hotspots for these and other motifs associated with chemical degradation does not mean a modification will always occur. Work probably too recent to have been cited in the book is beginning to provide actual data around deamidation and isomerization in sites within antibody variable domains. Signal peptides can also have an effect on productivity and output quality, and software tools are indicated that could be useful to predict cleavage points. As in many other aspects, these predictions need to be confirmed by experimental approaches like mass spectrometry. The authors close the chapter emphasizing the limitations of current computational tools to predict most issues; however, they are optimistic that the vast amount of experimental data being generated and collected will set the stage in the not too-distant future further to enable predictive approaches.

Almagro and Mascioni (Pfizer) in Chapter Nine, highlight the relatively recent paradigm shift to apply knowledge-based design principles and experimental screening to select favorable molecules early in the process. This is in contrast with the idea of trying to fix, by engineering or additional process development, “badly behaved” leads. The authors indicate how developability assessment (DA) may start at the “pre-discovery” stage by applying appropriate principles and knowledge at the library design stage, when dealing with synthetic antibody repertoires, or at the initial phases of optimization when designing, for example, affinity maturation libraries around an initial lead of “natural” or synthetic origin. There seems to be a more active role in the described work flow for explicitly predictive tools like the developability index (DI) described earlier in the book. Estimation of DI necessitates a 3D model of the Fv, at least, and the authors, who have been involved in this important line of research, describe some of the current approaches, issues and limitations for this requirement. Once again, the potential for predictive tools, given their speed and low cost, is emphasized at the conclusion of the chapter. A possible example of how this vision may be beginning to take shape is described in very recent work by Sharma and co-workers.3

The book closes with Chapter Ten by Zurdo, Arnell, Obrezanova, Smith, Gallagher, Gómez de la Cuesta and Locwin (Lonza). This is a substantial contribution, with close to 70 pages it is easily the longest chapter, and brings a perspective distinct from the previous 3 chapters authored by scientists in companies that most often discover and develop their own drugs. While a fair account of specific experimental and predictive tools is given, these tend to complement what has been offered in earlier chapters. A more unique aspect covered here is how one manages the process in the context of more formal quality-by-design and risk awareness literature. The authors’ perspective is that DA is not just about listing individual issues or problems in each therapeutic candidate, but about managing risk by pointing to the most significant failure points and then implementing appropriate control and corrective measures to address those key risk issues. Several decision-making tools are described that at the very least open the reader’s perspective to areas of knowledge quite distinct from the typical biophysical engineer’s “library.” Another interesting point of discussion in this chapter is the concept and application of surrogate assays or analytical methodologies whereby metrics that require complex procedures and large amounts of material may be replaced or approximated by measurements of related physical properties. Specific examples include use of apparent hydrodynamic radius by means of dynamic light scattering as a surrogate for viscosity, and high-throughput self-interaction chromatography as a surrogate for solubility. Such surrogate assays can be a powerful tool in the early stages of a discovery project, when many candidates need to be evaluated and rank-ordered.

In summary, this book represents a compelling opening salvo to the field of computational methods applied to antibody developability. The chapters are all generally well written and illustrated, and supported by extensive and timely literature citations: the average number of citations per chapter goes above 90, with over 50% of the references dating to 2009 or later. Developability of Biotherapeutics: Computational Approaches should be a good addition to the libraries of industrial and academic groups engaged in, or supporting, development of biopharmaceutical drugs.

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No potential conflicts of interest were disclosed.

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References

1. Developability of Biotherapeutics: Computational Approaches. Kumar S, Singh SK, editors. Boca Raton (FL): CRC Press; 2015.
2. Sydow JF, Lipsmeier F, Larraillet V, Hilger M, Mautz B, Molhoj M, Kuentzer J, Klostermann S, Schoch J, Voelger HR, et al. Structure-based prediction of asparagine and aspartate degradation sites in antibody variable regions. PLoS One 2014; 9:e100736; http://dx.doi.org/10.1371/journal.pone.0100736
3. Sharma VK, Patapoff TW, Kabakoff B, Pai S, Hilario E, Zhang B, Li C, Borisov O, Kelley RF, Chorny I, et al In silico selection of therapeutic antibodies for development: viscosity, clearance, and chemical stability. Proc Natl Acad Sci U S A 2014; 111:18601-6; PMID:25512516; http://dx.doi.org/10.1073/pnas.1421779112