

PERSPECTIVE

Recruiting the Immune System Against Disease: Lessons for Clinical and Systems Pharmacology

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Exposure–response analyses based on pharmacokinetics (PK) and pharmacodynamics (PD) offer great promise in research and development for vaccination and immunotherapy. While clinical and systems pharmacology integrate actionable multiscale information, quantitative immunology has focused on structural models of immune responses without data-based calibration or prediction. Given the growing immune data sets and to facilitate a systems approach to immunomodulation, we propose a paradigm shift in which the immune response, rather than the PK, captures “exposure.”

CLINICAL PHARMACOLOGY PRINCIPLES FOR IMMUNOMODULATION

Immunomodulation differs from other pharmacological interventions. First, it is characterized by the delayed emergence of immune responses caused by, e.g., the slow maturation of antibodies following vaccination or the emergence of T-cell responses after immune checkpoint inhibition. Although biological delays are not unique to immunotherapy and have been well characterized by the traditional pharmacokinetic-pharmacodynamic (PK-PD) paradigm, additional value lies in understanding the specific mechanisms by which an immunomodulator activates (or inhibits) the immune system, which do not only relate to target turnover or physical drug distribution. Second, the immunomodulatory response is persistent, often lasting much longer than the original intervention because of memory cells that preserve information arising from the antigenic challenge or immune checkpoint inhibition enabling the activation of exhausted T cells. Lastly, these responses can functionally differ between (apparently) similar interventions, such as when modestly different vaccination doses or schedules give rise to profoundly different humoral immune responses or tumor-infiltrating leukocytes lose function as a result of unfavorable microenvironment signals.

Therapeutic approaches directed at modulating immune responses do not easily fit in customary clinical pharmacology paradigms. Stroh *et al.*¹ first emphasized the need for mechanistic models (conceptual and computational) to interpret efficacy data in immune-competent, nonclinical model systems. Second, strong synergy between modeling and simulation and program acceleration is needed. Third is the

recommended phase II/phase III dose: for any therapeutic, optimizing dose, infusion duration and schedule requires understanding dose–exposure–response relationships. Lastly, effective model-informed drug development requires realistic models that go beyond empirical delays and explicitly represent effector immune system components.

The need to reframe dose–exposure–response paradigms for immunomodulatory interventions

One of the defining characteristics of immunomodulatory therapeutics is that the PD effect on disease is not directly because of administered therapeutics but because of immune response mediators that are modulated following a dosing event. This has been recognized by others: in inflammation,² indirect drug effects are on immune cell responses, immune cells “mediate” inflammation, and the causal cascade goes from PK (drug exposure) on to PD (effect on mediators) and then to response against a certain disease (i.e., prevention of tissue damage).

Traditional PK-PD constructs do not straightforwardly apply to immunomodulation. For example, in therapeutic vaccination there is no “dose” that directly relates to response as the half-life of vaccines is relatively short and is not predictive of the resulting long-lived immune response. Hence, “exposure” as the mediator of effect needs to be defined: does it relate to the therapeutic agent (e.g., vaccine or immune checkpoint inhibitor) or the effectors (e.g., the secreted immunoglobulins following vaccination or the increase in activated T cells postcheckpoint inhibitor treatment), which are slow to emerge and mature and somewhat distant from the dosing event(s)? As proposed to improve the probability of demonstrating an efficacy benefit, the framework of the “3 pillars of survival,”³ which include exposure at the target site of action (pillar 1), binding to the pharmacological target (pillar 2), and expression of pharmacological activity (pillar 3), is ill suited for immunotherapy. For reasons we have already mentioned, the separation between pillars 2 and 3 can be profound.

Adding to this already complex picture, immune response markers are not necessarily reflective of PD: strong immune responses can arise that yield poor or no efficacy. The experience with CYT006-AngQB (Cytos Biotechnology, Schlieren, Switzerland), a therapeutic vaccine against angiotensin II studied for the treatment of high blood pressure, demonstrates the importance of vaccination schedule and quality

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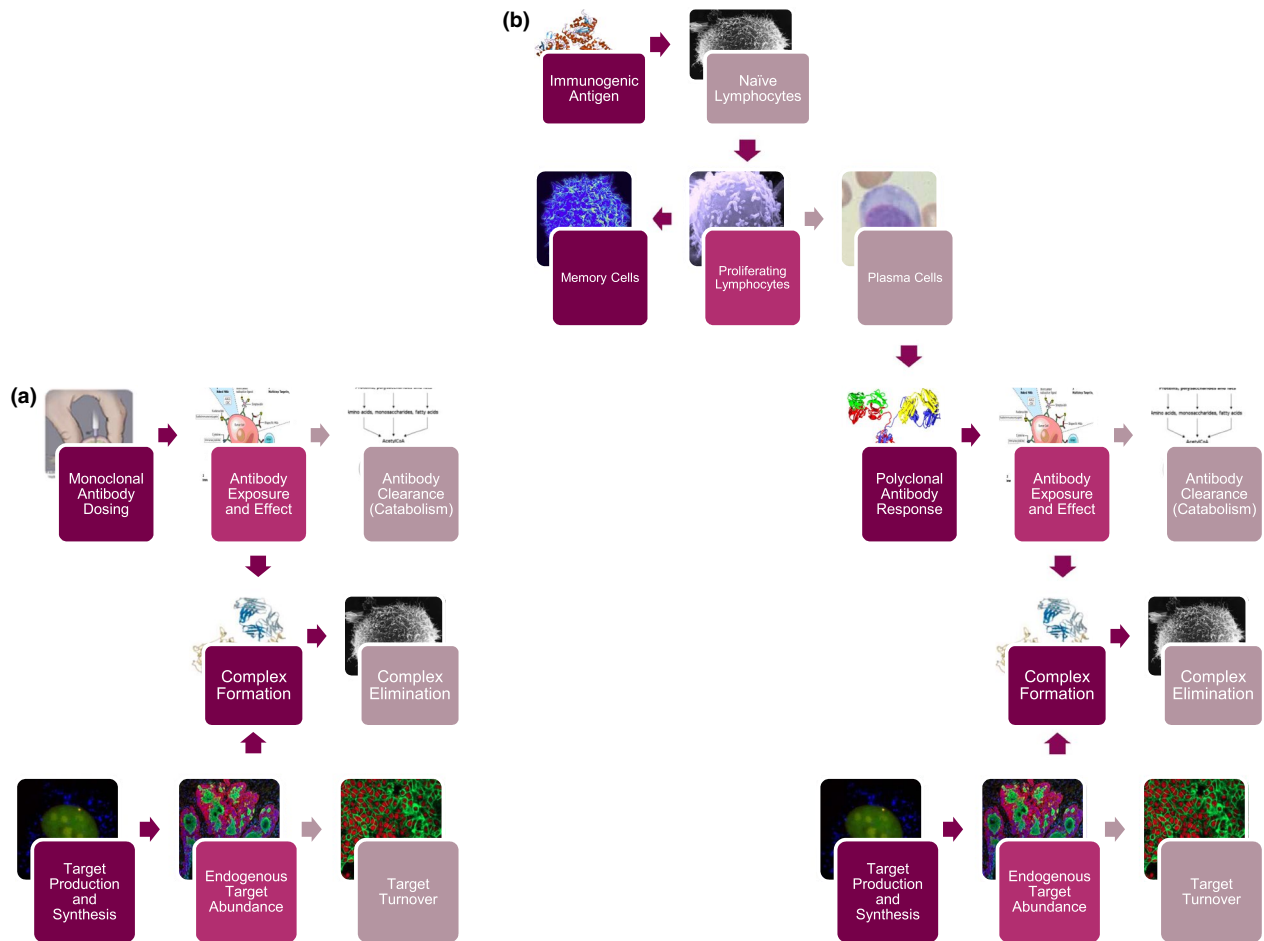


Figure 1 The cascade of events following subcutaneous dosing of a monoclonal antibody (**a, left**) is compared with that following therapeutic vaccination for eliciting a humoral immune response (**b, right**). In **a**, the monoclonal antibody is subject to distribution and clearance, and it also binds to the intended target, which has its own turnover characteristics. The antibody–target complex is then cleared according to its own mechanism. In **b**, a polyclonal antibody response is generated following vaccination with a suitable antigen and adjuvant. The immunogenic antigen stimulates naïve lymphocytes that, following clonal selection, differentiate in proliferating lymphocytes and plasma cells. Plasma cells secrete immunoglobulins that are polyclonal and essentially a circulating version of the B-cell receptors. Apart from its polyclonality, once the antibody response encounters the target, it resembles the fate of a monoclonal antibody. Image credits: Monoclonal antibody response: image from National Institutes of Health Medical Arts, https://www.cc.nih.gov/ccc/patient_education/pepubs/subq.pdf (public domain).

Antibody exposure and effect: image from https://commons.wikimedia.org/wiki/File:Monoclonal_antibodies.svg (public domain).

Antibody clearance (catabolism): image from TimVickers, <https://commons.wikimedia.org/wiki/File:Catabolism.svg> (public domain).

Complex formation: image from Fvasconcellos, https://commons.wikimedia.org/wiki/File:Trastuzumab_Fab-HER2_complex_1N8Z.png (public domain).

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Memory cells: image from National Institute of Allergy and Infectious Diseases, <https://www.flickr.com/photos/niaid/5950870236/> (licensed under CC BY 2.0).

Proliferating lymphocytes: image from Dr Triche, National Cancer Institute, <https://visualsonline.cancer.gov/details.cfm?imageid=1944> (public domain).

Plasma cells: image from <https://commons.wikimedia.org/wiki/File:Plasmacell.jpg> (public domain).

Polyclonal antibody response: image from Tim Vickers, https://commons.wikimedia.org/wiki/File:Antibody_IgG2.png (public domain).

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Endogenous target abundance: image from Markus Schober and Elaine Fuchs, The Rockefeller University, New York, NY, <https://visualsonline.cancer.gov/details.cfm?imageid=9852> (public domain).

Table 1 Specific examples of immune responses and biomarkers in various immunotherapy contexts

Category and example class of drug	Immune response mediator	Accessible biomarker distal to patient response	Accessible biomarker proximal to patient response
Vaccination Influenza vaccine	Neutralizing antibody titer and memory T cells/naive B cells	Circulating antinfluenza MP/HA/NA antibodies Matrix Protein, Hemagglutinin, Neuraminidase	Protection/immunity to infection
Suppression Anti-TNF Tumor Necrosis Factor	TNF (cytokine)	Reduction in free (non-drug bound) TNF levels	Disease scores—ACR American College of Rheumatology (arthritis) or PASI Psoriasis Area and Severity Index (psoriasis)
Suppression Peptide immunotherapy	Dendritic cells/T cells	Expansion of tolerogenic T cells, loss of <i>ex vivo</i> responsiveness to antigen	Reduction in effector T cells or cytokine release following challenge
Activation Anti-PD-1 Programmed Death-1	Activated, tumor-specific T cells	Circulating cytotoxic T-cell activation and proliferation	Antigen-specific tumor infiltrating lymphocytes

of raised immune response. In an initial phase II study,⁴ CYT006-AngQB was dosed at 300 and 100 µg at weeks 0, 4, and 12, providing antibody titers that resulted in ambulatory blood pressure lowering at the higher dose. To increase titers, in a subsequent study, the vaccine was given at weeks 0, 2, 4, 6, and 10. However, this new schedule, although providing high titers, failed to produce a blood pressure effect. This is likely because of the more frequent vaccine schedule eliciting immunoglobulins with decreased affinities.⁵ The antibody response observed after vaccination is usually polyclonal, i.e., displays a broad spectrum of dissociation constants and concentrations as was recognized in the early days of immune response modeling.⁶ Assessment via titers (e.g., using Enzyme-Linked Immunosorbent Assays), which represent the highest dilution at which an effect is observed, masks the relationship between immunoglobulin concentrations and affinities, with the titers being a mix of both. In other words, a high titer may not correspond to antibodies with the desired affinity, and higher or more frequent vaccine doses do not consistently yield an increase in the quality of the immune response. This of course differs from passive immunotherapy, in which a monoclonal antibody is manufactured to have a single affinity constant (**Figure 1a**). Postvaccination antibodies are instead polyclonal, show a range of immune responses, and are “produced” inside the body (**Figure 1b**), with the vaccine essentially providing “manufacturing instructions” to the host’s B cells. A quantitative pharmacology model that would account for the observed differences in the CYT006-AngQB response would likely have to incorporate affinity maturation and note how affinity maturation of the polyclonal response is influenced by dosing schedule for the specific construct under consideration. It would be difficult to predict the behavior of the two schedules based on typical PK-PD approaches because a higher level of mechanistic detail is required. This kind of complexity extends to both humoral and cellular responses and points to the realization that raised immune responses are more akin to PK (exposure) than PD.

These considerations lead to a proposal to reframe the dose–exposure–response framework for the study of immunomodulatory interventions (**Figure S1**). We propose to consider, as the equivalent to exposure, the temporal profile and characteristics of mediators (immune cells or antibodies)

elicited by the immunomodulator, as opposed to drug concentration or summary PK parameters (peak concentration, area under the curve, etc.). The equivalent of *dose* would be the immunomodulator dosing time or concentration–time course at the site of drug action, as opposed to the customary amount of drug administered, infusion rate, dosing schedule. The equivalent of *response* would not change and remain a suitable biomarker proximal or distal to, but always correlated with, patient response (e.g., blood pressure in the CYT006-AngQB example). By shifting the emphasis on the raised immune response, we focus our attention on the true mediators of PD and avoid the potential confusion generated by exclusively optimizing humoral and cellular responses as opposed to biomarkers representative of the desired effect. Examples of this shift are presented in **Table 1** to further clarify our thinking.

This reframing has implications for bioanalytical sciences. Simply written, the discipline of clinical pharmacology needs to pursue quantification of immune response correlates with the same vigor as it has pursued quantification of drug exposure. In the case of the humoral (immunoglobulin) immune responses, these correlates include production of antibody by plasma cells and their affinity ranges and kinetics (ideally, concentrations rather than titer). The monitoring and optimization of affinity maturation would prevent the emergence of ineffective responses and would allow discrimination among dosing schedules. In other words, assays need to be designed to properly quantify the immune system components that are specific to the target antigen. For the cellular immune response, methods to monitor it *ex vivo* (both peripheral and tissue) are readily available, e.g., Enzyme-Linked Immunosorbent Assays and flow cytometry. However, it is of paramount importance to monitor antigen-specific cellular responses relevant to the intended indication because these are more likely to represent a true PD effect, i.e., one coupled with improved clinical efficacy. This was demonstrated in a study of non-small cell lung cancer patients that correlated tumor antigen burden and subsequent prevalence of tumor antigen-specific T cells with durable responses to immune checkpoint blockade.⁷ Cell migration and tissue infiltration would also be important to quantify (the equivalent of systemic and site of action exposures in

the traditional setting), and novel image analysis strategies could be used to better characterize *in situ* immune correlates.⁸

PK-PD modeling and simulation, currently a mainstay of clinical pharmacology, can contribute to the optimization of immunomodulation. Parsimonious PK-PD approaches account for minimally required features of the immune response: timing (schedule) of immunotherapy administration and the resultant time course of immune response mediator(s), partitioning of the target population between responders and nonresponders (by mixture statistical models), and counter-regulatory response (resistance or immune regulation, e.g., by regulatory T cells). PK-PD can substantially benefit from more realistic systems pharmacology approaches^{9,10} that elucidate the mechanism, timing, and extent of emerging immune responses depending on the questions posed by the drug discovery and development team.

Ultimately, these considerations can have an impact on experimental and trial design and perhaps on drug approval and clinical practice. We do not intend to provide guidance for how to capture this framework in a drug label, although we can certainly anticipate an evolution in immunotherapy toward a more personalized approach that could well require additional descriptors of the immune response in the label. There is likely value in some real-time monitoring to adjust dosing (level and/or frequency) to enable a successful outcome for patients. Specific parameters and how to monitor them will depend on each drug. Such companion “diagnostics” may not be cheap to develop and implement and would only be viable if they brought added value to patient survival and quality of life. Personalized medicine is certainly on the horizon, and we believe that the paradigm shift proposed here will accelerate our understanding of how to implement personalized medicine strategies for immunomodulatory drugs.

Immunology is often an empirical science. Immunomodulatory approaches can bring effective and durable interventions: the challenge of what to measure and when is compounded by the complexity of raised immune responses. We suggested here a reframing of dose–exposure–response science for immunomodulation. Our purpose is to frame the question as clearly as possible, and it is our hope that this reframing will improve communication between clinical pharmacologists and experimental immunologists and suggest both avenues for data collection and ideas for experimental design.

Supporting Information. Supplementary information accompanies this paper on the *CPT: Pharmacometrics & Systems Pharmacology* website (www.psp-journal.com).

Figure S1. A view of the traditional dose-exposure-response paradigm is compared with modifications required to accommodate the unique features of immunomodulation.

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