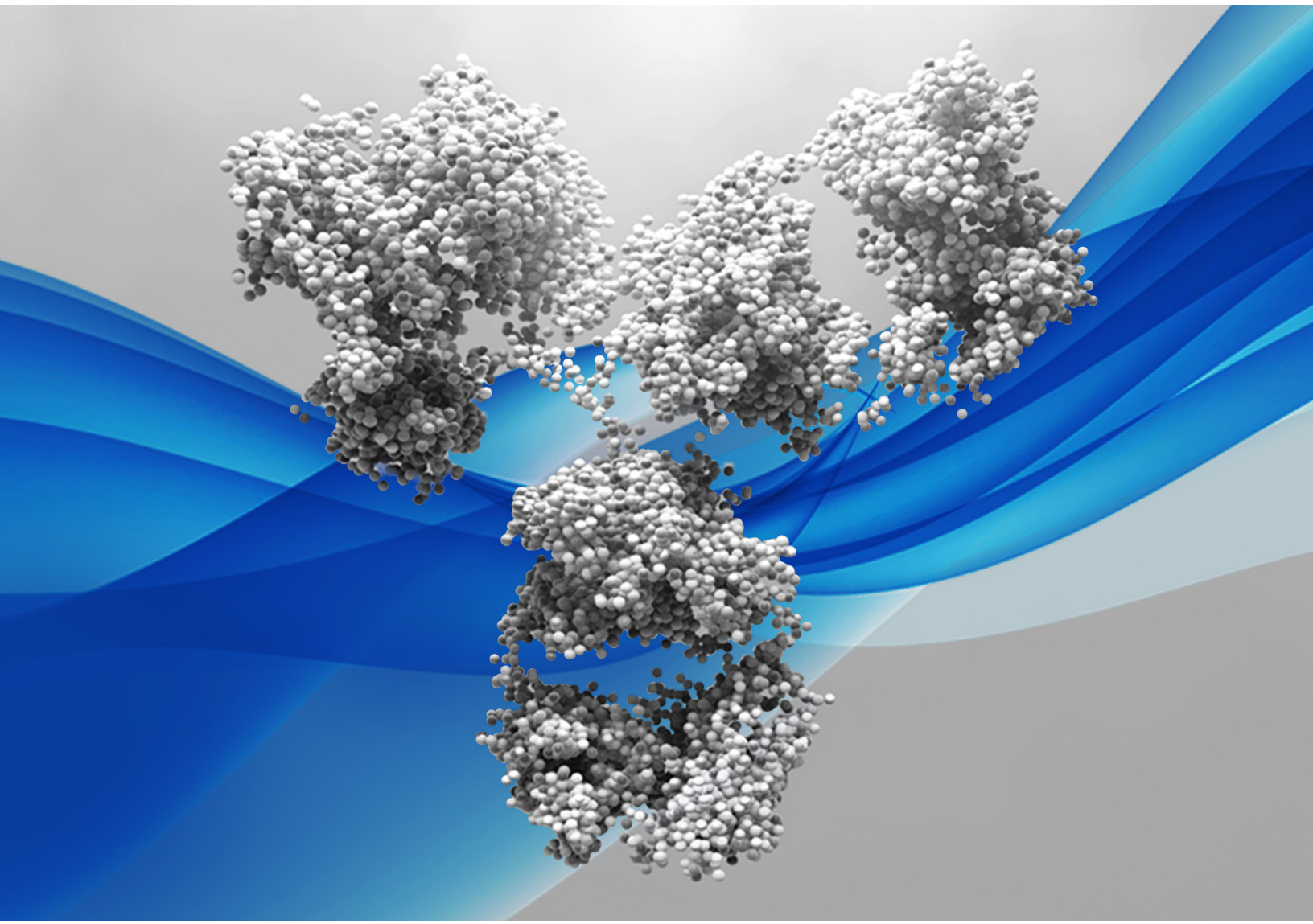
药明康德
WuXi AppTec



The 2019 FDA Guidance's Implications for Immunogenicity Assessment

In early 2019, the FDA updated its guidelines for immunogenicity assessment. The guidance further clarifies FDA's recommendation when compared to the previous guidelines published in 2016. In this white paper, we document some of the significant changes and discuss the implications on drug development programs.

What's Different in 2019?

Many aspects of this guidance were revised, but the significant revisions focus on the following areas:

- Risk assessment
- Statistical approaches to ascertain cut point
- Removal of long-term stability requirement
- Minimal required dilution
- Positive control antibodies
- Development of assays to measure neutralizing antibodies
- Strategies for managing pre-existing antibodies
- Updates in documentation requirements

Risk Assessment

In the 2019 guidance, the FDA recommends that immunogenicity risk assessment and rationale should be provided prior to entering first in human (FIH) studies and submitted in conjunction with the IND. Previously, the FDA advised that sponsors should provide a rationale for immunogenicity testing rather than the risk assessment itself. The FDA also provided clarification that test samples during phase 1 and phase 2 studies should use suitable screening, confirmatory assays, and, where necessary, neutralization assays. Previously, the FDA was less explicit about the types of testing that should be done during phase 1 and phase 2 studies.

Following risk assessment and general approach of the multi-tiered strategy, the new guidance provides much more clarity and instructions on how to deal with "domain specificity". The recommendations are that initial screening and confirmatory assays are designed to detect antibodies against the whole therapeutic protein product. For confirmed positive samples against the whole molecule, examination of immune responses to specific functional domains may require that sponsors develop domain specific assays.

When immunogenicity poses high clinical risks, the 2019 guidance also recommends development of all immunogenicity assays, which may include NAb assays, for their intended purposes before moving into FIH, and sample testing being performed in z real time.

Statistical Approach Update

The cut point of the assay determines whether the sample response is positive or negative; establishing the appropriate cut point is crucial to reducing false negative results. Assays should be designed such that they generate a 5% false positive rate, which is important for ensuring the assay identifies all subjects who may develop antibodies to the therapeutic protein.

Historically, the assay cut point was determined directly from a normal percentile, which assumes that distribution of results will follow a traditional bell-shaped curve. This method is simple, but it significantly underestimates the false positive rate, which results in a lower chance of satisfying the 5% false positive rate. For example, the traditional method for screening assays is estimated to produce a cut point only having less than a 50% chance to satisfy the required 5% false positive rate.

In the 2019 FDA guidance, the FDA requires sponsors to find a statistically sound method to determine the cut point, whereas previous guidance documents stated that cut point estimation could be achieved with a small number of samples. The 2019 guidance offers an approach wherein a screening assay should have at least a 90% chance to satisfy the 5% false positive rate and a confirmatory assay should have at least an 80% chance to satisfy the 1% false positive rate. The assay sensitivity can be calculated by interpolating the linear portion of the dilution curve to the assay cut point. And low positive control, as an important system suitability control, should

[1] M. Shen, et. al., J Biopharm Stat, 25(2): 269-279 (2015).

be set at a level that is consistently above the cut point with a targeted 1% failure rate. Statistical analysis can add value in determination of assay sensitivity and low positive control.

This is not a generally accepted standard method for these statistical determinations. One way is a statistically sound estimation package including an outlier exclusion and cut point estimation to satisfy the new FDA requirements. The statistical methodology is based on order statistics, Bayesian, and Monte Carlo methods and can be applied to any assay data given that there are at least 50 samples. These methods are designed to satisfy or exceed the FDA confidence requirements without being too conservative. The required computations have been implemented using the standard statistical programming languages SAS and R.

Removal of Long-term Stability Requirement

The new guidance removes the expectation of long-term stability testing on positive control antibodies, as it is well known that antibodies are generally stable under -70°C for years and that in immunogenicity assessment, a surrogate positive control antibody is used for long-term stability assessment which does not reflect the clinical sample stability. Instead, the guidance now advises that sponsors minimize freeze-thaw cycles by appropriately aliquoting subjects' samples and evaluating short-term stability. This includes freeze-thaw cycle and refrigerator- and room-temperature stability of positive control antibodies.

Minimal Required Dilution (MRD)

Matrix components are known to interfere with assay selectivity and contribute to non-specific signals and potentially obscure positive results. The FDA guidance in 2019 allows sponsors to use one of three definitions of the MRD –

- Sample dilution that yields to highest signal-to-noise ratio
- Sample dilution that results in a signal closest to assay diluent
- Sample dilution that results in the highest signal to variability ratio

The expansion of MRD definitions allows sponsors more options to achieve optimal selectivity for an assay.

Positive Control Antibodies

Compared with the 2016 guidance, the 2019 guidance adds clarification and detail about utilizing the positive control antibody to evaluate system suitability, including the following detailed new language:

Once a source of a positive control antibody has been identified, it should be used to assess assay performance characteristics such as sensitivity, selectivity, specificity, drug tolerance, and reproducibility. FDA recommends that the positive control antibody should be reserved for use as a quality control or system suitability control during routine performance of the assay. For assay development and validation, dilutions should generate high, intermediate, and low assay signal values. The intermediate value is useful for assessing precision during assay validation. This is recommended even for development of qualitative assays to understand whether assay performance is acceptable across a broad range of antibody concentrations. Intermediate-value QC samples for detection of ADA are generally not needed for monitoring system suitability during routine assay performance.

Neutralizing Antibodies

Neutralizing antibodies (NAbs) can have a significant impact on drug PK, PD, safety and efficacy, and the overall impact of antidrug antibodies may correlate with the activity of NAbs. The FDA has made extensive updates to its guidance for assays on NAbs' action. The FDA has increased its flexibility in terms of the type of assay that can be used to assess NAbs. The FDA explicitly allows utilization of either a highly sensitive PD marker, properly designed PK assay, or both to generate data capable of informing clinical activity, in lieu of a NAb assay. Cell-based NAb assays are notoriously insensitive and highly variable, and they may not be as informative as a suitable PD or PK assay to indicate neutralizing antibody activities.

Pre-Existing Antibodies

PEGylation is a common modification of certain proteins that can reduce the immunogenicity and prolong the circulatory life of a protein or, improve water solubility of certain proteins. However, patients may have pre-existing antibodies against PEG, given its frequent use in products such as cosmetics.

The fact that patients entering clinical trials may already have pre-existing antibodies challenges the statistical analysis traditionally used to differentiate between positive and negative assay results. The challenge with pre-existing antibodies is determining the cut point that adequately differentiates negative ADA, pre-existing antibodies and true ADA positive samples. The 2019 guidance provides more details on how to manage the statistical calculations on the cut point during method validation, as well as, discussing reporting strategies for capturing data around pre-existing antibodies. It gives a clear definition of treatment-boosted ADAs when there are pre-existing antibodies present and the titer of antibodies increases after exposure to the therapeutic protein versus differentiating them from treatment-induced antibody titers.

Documentation Requirements

Prior to the 2019 guidance, immunogenicity data was dispersed throughout the electronic common technical document (eCTD), the standard format for submitting to the FDA. This presented challenges for reviewers to understand the big picture of a potential therapeutic protein's immunogenicity profile. The 2019 guidance provides detailed direction on documenting immunogenicity and requires an integrated immunogenicity summary report that provides a clear overview to the FDA reviewers to help understand the immunogenicity data up front. In addition, the FDA recommends sponsors to arrange the integrated summary into distinct sections.

Documentation Requirements

1. Immunogenicity Risk Assessment
2. Tiered Bioanalytical Strategy and Assay Validation Summaries
3. Clinical Study Design and Detailed Immunogenicity Sampling Plans
4. Clinical Immunogenicity Data Analysis
5. Conclusions and Risk Evaluation and Mitigation Strategies (REMS)

Conclusions

The 2019 FDA guidance on immunogenicity assessment helps to understand the potential immune responses that patients may encounter when exposed to a therapeutic protein. The updates are complex and it can be difficult to determine the right assays for your IND- and BLA-enabling studies. A trusted partner who has the scientific and regulatory expertise to help ensure the development plan of your biologic drug meets the changing regulatory requirements. WuXi AppTec can collaborate on your program and our team of experts can help address these changes in the design and execution of bioanalytical testing needed for submission success.

药明康德
WuXi AppTec

Contact Us to Learn More

labtesting.wuxiapptec.com | info_ltd@wuxiapptec.com

NEW JERSEY | SHANGHAI | NANJING | SUZHOU | ATLANTA | ST. PAUL