Abstract

Purpose: To develop a strategy for optimizing sample pretreatment steps to overcome drug interference in anti-drug antibody (ADA) assays for therapeutic biologics including antibody drug conjugates (ADCs).

Background: Biologics commonly elicit immune responses, resulting in ADA-activated drug dissociation (the presence of drug can cause a variety of clinical consequences ranging from relatively mild to serious adverse events). ADA assay drug tolerance, antibodies, and other methods can be used for ADA removal. However, the presence of high drug levels in clinical samples can interfere with detection of ADA using LBA by masking epitopes necessary for the assay. A critical step in this process is to develop a standard method to isolate ADCs prior to enzyme-linked immunosorbent assay (ELISA) or related methods. This method was designed to use an antibody-therapeutic conjugate to improve ADA detection efficiency.

Methods: We describe a step-wise approach to optimize the acid dissociation pre-treatment step coupled with an ELISA-based bridging assay system for an antigen-antibody therapeutic.

Results: We confirmed that low pH was detrimental to ADA activity and that effective neutralization of the acid was essential to preserve maximum ADA activity. All components of the assay were systematically optimized to achieve high ADA sensitivity (0.01 ng/mL) and maximum signal (3000:1). Moreover, significant improvements in drug tolerance were observed ranging from 20-100 µg/mL depending on the assay system. Overall, treatment improved drug tolerance significantly (3-4 fold) and would be expected for all clinical trials.

Conclusions: This approach was successfully applied to develop, validate, and improve ADA assays for multiple biologics including antibody drug conjugates. Pre-treatment of samples significantly improved the drug tolerance and the assay sensitivity for the ADA detection in human serum for multiple biologics.

Step-wise Approach for Minimizing Drug Interference in Immunogenicity Testing for Therapeutic Biologics

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Biologic drugs commonly elicit immune responses, resulting in anti-drug antibody (ADA) that can cause a variety of clinical consequences ranging from relatively mild to serious adverse events. ADA assay drug tolerance, antibodies, and other methods can be used for ADA removal. However, the presence of high drug levels in clinical samples can interfere with detection of ADA using LBA by masking epitopes necessary for the assay. A critical step in this process is to develop a standard method to isolate ADCs prior to enzyme-linked immunosorbent assay (ELISA) or related methods. This method was designed to use an antibody-therapeutic conjugate to improve ADA detection efficiency.

Methods

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References


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