Perpetrator or Victim: Evaluating Therapeutic Proteins in Drug-Drug Interaction Studies

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Unlike many small molecules, therapeutic proteins (TPs) are not metabolized by CYP450, but are rather cleared by renal filtration, protease degradation or binding to receptors. As a consequence, the pharmacokinetics (PK) of TPs are less likely to be impacted by concomitantly dosed drugs that have potential to induce or inhibit CYP450 enzyme activity. However, by up- or down regulating the expression of CYP450 enzymes, TPs may alter the PK of other co-administered drugs. Moreover, by affecting physiological processes, some TPs may interfere with the pharmacodynamic (PD) effect of concomitant drugs [1, 2].

In August 2020, the FDA issued **draft guidance** to assess potential interactions between TPs and other drugs [3]. While the guidance applies to TPs, general concepts can similarly extend to other biological products such as cellular and gene therapies.

Key elements in this new guidance were previously covered by the draft Guidance for Industry from 2012, entitled “Drug Interaction Studies - Study Design, Data Analysis, Implications for Dosing, and Labeling Recommendations”. However, neither the subsequent draft issued in 2017 (“Clinical Drug Interaction Studies - Study Design, Data Analysis and Clinical Implications”) nor the (final) guidance entitled “Clinical Drug Interaction Studies - Cytochrome P450 Enzyme- and Transporter-Mediated Drug Interactions,” issued in early 2020, addresses DDIs involving TPs [4].

Therapeutic proteins have a different interaction profile compared to small molecules and in keeping with the perspective of steadily increasing numbers of biologicals in development and reaching the clinic, the new, standalone (draft) FDA guidance for TPs provides relevant directions towards considering drug-drug interaction (DDI) studies and their design, specifically for the development of TPs. This guidance also includes a decision tree that provides a pragmatic approach to evaluate the need for DDI studies [3].

The classification of TPs and their potential for interaction with other drugs is summarized in the table hereafter, including examples for each category.

Table 1. Summary of When to Conduct a Therapeutic Protein DDI Study

Therapeutic Protein	When to Conduct a DDI Study	Scenario Example	Study Design
Proinflammatory Cytokine-Related Mechanisms			
Proinflammatory cytokine	Evaluate DDI potential	Peginterferon	Evaluate TP as a perpetrator
Cytokine modulator (increases proinflammatory cytokine levels)	Determine DDI potential, if probable conduct study. If DDI potential is low contact FDA for guidance and provide justification	Somatropin (potential to increase cytokine expression) [5]	Evaluate TP as a perpetrator

Therapeutic Protein	When to Conduct a DDI Study	Scenario Example	Study Design
Cytokine modulator (decreases proinflammatory cytokine levels)	No study required, but label should include potential for DDI. If potential for DDI is low, justification is required. Alternatively, conduct DDI evaluation	IL-6 and TNFα inhibitors (e.g. tocilizumab, golimumab – see text)	
DDIs Unrelated to Proinflammatory Cytokines			
Affects physiological process that can alter PK profile with drug co-administration	Evaluate DDI potential	GLP-1 receptor agonists (delayed gastric emptying)	Evaluate TP as a perpetrator
Drug co-administration affects TP disposition	Evaluate DDI potential	Statins and fibrates (increased disposition of alirocumab and evolocumab – see text)	Evaluate TP either as a perpetrator or victim
FcRn co-administration can affect TP	Evaluate DDI potential	Blocking between TP containing Fc region of human IgG and FcRn	Evaluate TP as a victim
Immunosuppressor co-administration with TP affected by immunogenicity	Evaluate DDI potential	Methotrexate (reduced clearance of infliximab, adalimumab – see text)	Evaluate TP as a victim
Antibody-Drug Conjugates			
Antibody-drug conjugates (ADC)	Evaluate DDI potential for both (free) small molecule drug and antibody component. Refer to [4].		Evaluate ADC as a victim if free small molecule concentration is too low.

Adapted from **Drug-Drug Interaction Assessment for Therapeutic Proteins Guidance for Industry** [3].

While in chronic inflammatory diseases circulating cytokines may induce upregulation of CYP enzyme activity, monoclonal antibodies (mAbs) like tocilizumab, an anti-IL-6 receptor antibody, may reverse the “inhibitory” effect of IL-6, thus changing CYP activities in patients with rheumatoid arthritis back towards normal levels [6].

Co-administered drugs may also affect the disposition of TPs; for instance, statins and fibrates induce PCSK9 expression, which subsequently leads to enhanced cellular uptake and clearance of anti-PCSK9 antibodies like alirocumab and evolocumab [6]. However, according to the respective drug labels, the impact of such concomitant medication has no clinical significance [7, 8]. Methotrexate reduces the clearance of infliximab, adalimumab and golimumab by inhibiting the formation of antibodies against the mAbs, yet no dose adjustments are required for the perpetrator nor the victim drug (reviewed in [1, 6]).

In vitro and preclinical studies can provide mechanistic understanding of TP DDI potential. If a clinical TP DDI investigation is deemed necessary, efficiencies can be gained with a healthy volunteer study if the drug has an acceptable safety profile. Similar to conventional DDI studies, engaging healthy volunteers allows for a single-site solution, swifter enrollment and less PK variability. Moreover, healthy subjects would avoid potentially induced CYP activity in patients associated with their inflammatory status. Furthermore, studies can use a parallel or crossover design, depending on the suspected mechanism of the DDI and the PK characteristics of the drugs. For example, a parallel design may be more applicable for TPs with a long half-life or to examine the impact of anti-drug-antibodies (ADA). Single sequence crossover design (substrate followed by the substrate plus the TP) can be used when evaluating the TP as a perpetrator. Moreover, a cocktail approach may be an efficient means of evaluating the DDI for TPs where multiple CYPs could be impacted.

Alternatively, TP drug interaction data may be obtained from nested population PK (PopPK) studies, which may waive the requirement for DDI studies for certain proinflammatory cytokines. In this option, sponsors must ensure appropriate data capture and PK sampling. In addition, physiological based PK (PBPK) may help understand the underlying mechanism of a TP DDI. For instance, recent trials to simulate the effect of IL-6 on CYP enzyme activities have been conducted with the aid of PBPK modeling [9].

As the current guidance for TPs is a draft document, the final version may be subject to modifications such as to address **industry comments**. Some of the concerns raised by industry stakeholders state that while the draft guidance highlights the impact of proinflammatory cytokines on CYP enzyme activity, it was noted that cytokines might similarly affect the expression of hepatic drug transporters, which should be taken into account too when assessing the potential for DDIs with a TP. Moreover, with respect to ADCs, it was questioned whether data from DDIs with an ADC could be extrapolated to ADCs sharing the same small molecule payload. A further suggestion was to indicate the potential application of monitoring the ratio of 4-beta hydroxy cholesterol to cholesterol as an endogenous biomarker for CYP3A4/5 activity.

Another important consideration for any DDI study is the bioanalytical analysis. Bioanalytical efficiencies can expedite TP DDI studies in three key ways. First, a bioanalytical lab **co-located** within the clinical pharmacology unit (CPU) accommodates immediate analysis for assays requiring fresh samples. Secondly, an extensive offering of **analytically validated assays** for CYP substrates can save time and money. Finally, a **tiered approach** to assay validation for proinflammatory cytokines and immune factors, utilizing the most sophisticated instrumentation, can be applied to evaluate the impact of the TPs on cytokine levels. While any bioanalytical assay that will confirm a FDA decision should be fully validated, the effect of a TP on cytokines as exploratory or secondary endpoints may not need to be so rigorously validated in this 'fit-for-purpose' approach, which may result in reduced method development costs.

Figure 1. Tiered Bioanalytical Assay Validation Approach

<table>
<tr><th>Tier 1:
Exploratory Validation</th><th>Tier 2:
Partial Validation</th><th>Tier 3:
Full Validation</th></tr>
<tr><td colspan="2">No FDA decision required</td><td>FDA decision required</td></tr>
<tr><td>Mechanism of action</td><td>Lead candidate efficacy and optimization</td><td rowspan="2">Phase 1 safety</td></tr>
<tr><td>Disease sample screening</td><td>Preclinical studies</td></tr>
<tr><td>Lead biomarker/target selection</td><td>Secondary endpoint in clinical trial</td><td>Phase 2/3 clinical trials</td></tr>
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References:
1. Huang SM, Zhao H, Lee JI, Reynolds K, Zhang L, Temple R et al. Therapeutic protein-drug interactions and implications for drug development. Clin Pharmacol Ther. 2010;87(4):497-503.
2. Mahmood I, Green MD. Drug Interaction Studies of Therapeutic Proteins or Monoclonal Antibodies. J Clin Pharmacol. 2007;47:1540-1554.
3. Drug-Drug Interaction Assessment for Therapeutic Proteins Guidance for Industry; Draft Guidance for Industry. 2020. https://www.fda.gov/regulatory-information/search-fda-guidance-documents/drug-drug-interaction-assessment-therapeutic-proteins-guidance-industry.
4. Clinical Drug Interaction Studies — Cytochrome P450 Enzyme- and Transporter-Mediated Drug Interactions Guidance for Industry. 2020. https://www.fda.gov/regulatory-information/search-fda-guidance-documents/clinical-drug-interaction-studies-cytochrome-p450-enzyme-and-transporter-mediated-drug-interactions.
5. Lee JI, Zhang L, Men AY, Kenna LA, Huang SM. CYP-mediated therapeutic protein-drug interactions: clinical findings, proposed mechanisms and regulatory implications. Clin Pharmacokinet. 2010;49(5):295-310.
6. Ferri N, Bellosta S, Baldessin L, Boccia D, Racagni G, Corsini A. Pharmacokinetics interactions of monoclonal antibodies. Pharmacol Res. 2016;111:592-9.
7. Evolucumab: Highlights of Prescribing Information. 2019. https://www.accessdata.fda.gov/drugsatfda_docs/label/2019/125522s022lbl.pdf.
8. Alirocumab: Highlights of Prescribing Information. 2020. https://www.accessdata.fda.gov/drugsatfda_docs/label/2020/125559s024lbl.pdf.
9. Xu Y, Hijazi Y, Wolf A, Wu B, Sun YN, Zhu M. Physiologically Based Pharmacokinetic Model to Assess the Influence of Blinatumomab-Mediated Cytokine Elevations on Cytochrome P450 Enzyme Activity. CPT Pharmacometrics Syst Pharmacol. 2015;4(9):507-15.