CMC Regulatory Considerations for Antibody-Drug Conjugates (ADCs) – From the Biologics Perspective

Lei Zhang, Ph.D.

Office of Product Quality Assessment III
Office of Pharmaceutical Quality
FDA

April 29, 2026


Everyone deserves confidence in their *next* dose of medicine.
Pharmaceutical quality assures the availability, safety, and efficacy of *every* dose.
FDA
www.fda.gov
2

Disclaimer

This presentation reflects the views of the author and should not be construed to represent FDA's views or policies.

Outline

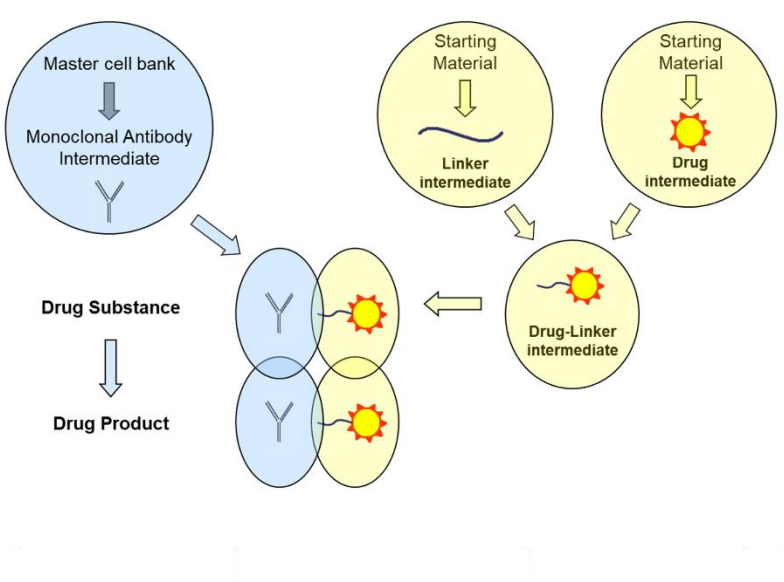
- ❑ Overview of the review process from the biologics perspective
- ❑ Expectations on ADC-specific characterization and control strategies
- ❑ Additional points to consider

Outline

- ❑ Overview of the review process from the biologics perspective
- ❑ Expectations on ADC-specific characterization and control strategies
- ❑ Additional points to consider

Biologics Review of ADCs

- Monoclonal antibody (mAb) as drug substance intermediate (DSI):
  - Expectations of the manufacturing process, characterization, and control strategy for the mAb intermediate are generally held to the same standards as for traditional mAbs.
- ADC drug substance (DS) and drug product (DP):
  - ADC characterization, DS and DP manufacturing processes, and the control strategies are reviewed from the biologics perspective.



Direct Control over the Manufacturing Process of the Biologics Component is Expected for BLAs

- Biologics License Applications and Master Files (Final Rule, 2/12/2024):
  - Applications for a biological product under the Public Health Service Act (PHS Act) may not rely on a master file (MF) for drug substance intermediate, drug substance, or drug product (DSI/DS/DP) information. (Exception: certain "deemed BLAs")
    - Applies to the biological product constituent part of a combination product (e.g., mAb intermediate for ADC).
    - Other types of cooperative manufacturing arrangements can be considered. See the 2008 guidance for industry “Cooperative Manufacturing Arrangements for Licensed Biologics”.
    - May rely on a MF for other kinds of information (e.g., excipients, stabilizers, penetrants, container closure, and other materials).

Outline

- ❑ Overview of the review process from the biologics perspective
- ❑ Expectations on ADC-specific characterization and control strategies
- ❑ Additional points to consider

ADC-Specific Characterization

- Identification of conjugation sites.
- Drug-to-antibody ratio (DAR) and drug load distribution (DLD).
- Impurities: unconjugated mAb and mAb associated process- and product-related impurities, small molecule associated impurities (e.g., free drug and drug-linker impurities), and conjugation reaction associated impurities (e.g., residual solvents and heavy metals).
- **Raw Materials used in the conjugation process.**
- **Impact of the conjugation process on the mAb intermediate.**
- **Biological activity.**

Raw Materials used in the conjugation process

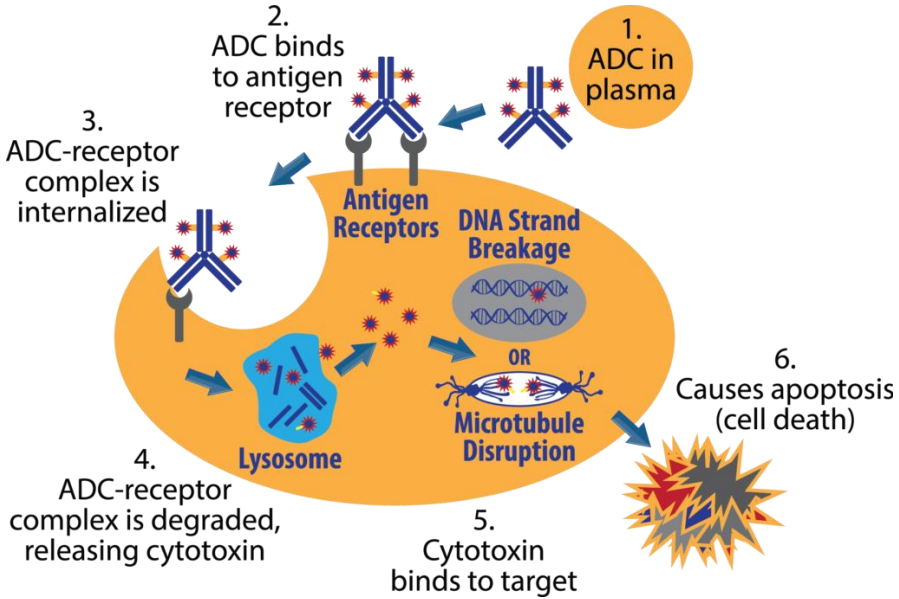
- **Safety and quality assessment of critical raw materials should be provided.**
  - e.g., enzyme(s) used for site-specific conjugation process in an ADC IND submission.
    - Origin of the DNA sequences coding for the enzyme(s).
    - Raw materials used for manufacturing the enzyme(s), including all animal-derived raw materials.
    - Safety and quality assessment of the enzyme(s), including viral clearance (if applicable).
    - An assessment of the capacity of the purification process for clearance of the enzyme(s). If needed, quantitative limits for residual enzyme(s) may be implemented for the release of clinical materials.
    - If the enzyme is considered high risk (e.g., with human endogenous counterpart), the sponsor may be requested to perform a risk assessment regarding the potential impact on immunogenicity.

Impact of the conjugation process on the mAb intermediate – Characterization Data to Support the Understanding

- Target binding:
  - ADC and mAb intermediate are expected to maintain similar binding properties and similar acceptance criteria for release and stability.
- Size variants:
  - Fragmentation and aggregation of the mAb intermediate compared to the ADC initially and during stability testing is expected to be assessed.
  - For hydrophobic drug-linkers, there may be an increase in aggregates compared to unconjugated mAb.
- Charge variants:
  - Chemical conjugation can impact the charge of the mAb intermediate. The resulting charge heterogeneity of ADCs can further impact product solubility, potency, and pharmacokinetics.
  - Charge based assays may not be possible/meaningful after conjugation to lysine residues.
- Effector function:
  - Fc-mediated effector functions and their link to the intended mechanism of action should be characterized. The purposeful disruption of inter-chain disulfide bonds from the hinge region of the mAb during site-specific or cystine-coupled conjugation may impact Fc-Fcγ receptor interactions and downstream effector functions (i.e., ADCC and CDC).

Biological Activity - Characterization

- Target binding assay: Demonstrates a critical step in the ADC mechanism of action (MOA).
- Cell-based cytotoxicity assay: Demonstrates the ADC MOA, including target binding, internalization, drug release, and cell killing.
- Bystander effect: If the bystander effect is a proposed MOA for an ADC, bystander effect activity should be characterized.
- Effector function: An appropriate control strategy for effector function (if present in MOA) should be justified with supporting characterization data.



https://njbio.com/antibody-drug-conjugates/

Considerations for ADC Specifications – What's Unique for ADCs

- DAR/DLD:
  - Appropriate methods should be used depending on the conjugation technology.
  - Acceptance criteria should provide adequate control of DAR to ensure safety and efficacy.
  - DLD may be included in the specification for site-specific conjugation.
- Potency:
  - Both target binding and cell-based cytotoxicity assays are generally expected.
- Impurities:
  - Appropriate upper limits are expected for product-related impurities (e.g., free drug and unconjugated mAb) and process-related impurities (e.g., linker impurities, solvent impurities, and heavy metals).
  - Additional process-related impurity limits (e.g., residual conjugation enzyme) may be needed depending on the conjugation process.
- Free thiols may be included for cysteine-engineered ADCs.

Outline

- ❑ Overview of the review process from the biologics perspective
- ❑ Expectations on ADC control strategies
- ❑ Additional points to consider

Additional Considerations for Lyophilized Drug Product (DP) Specifications

- Lyophilized DP:
  - uniformity,
  - moisture content,
  - reconstitution time,
  - gross content in “mg per vial”,
  - appearance, color, etc.

- Reconstituted DP:
  - Appearance, including visible particles,
  - color, clarity,
  - deliverable volume,
  - subvisible particles,
  - protein concentration, etc.

Compatibility and In-Use Stability Studies

- Simulate the reconstitution, dilution and administration of the product with multiple drug concentrations bracketing those dosages described in the clinical protocols (IND) or package inserts (BLA).
- Capture the proposed reconstitution solution(s) and diluent(s), and all potential product-contacting surfaces of commonly used administration material and devices, e.g., infusion set, in-line filter, syringes, closed system drug-transfer device (CSTD).
- Take samples from material that would be delivered to patients (e.g., the needle tip)
- Perform under real-use or worst-case conditions for storage duration, infusion rates, temperature, light, etc.
- Quality attributes to be tested: protein concentration, aggregates, visible and subvisible particles, potency, **change in DAR and/or the amount of free drug**, etc.
- In-use storage time should ensure microbial safety.

Other Considerations

- Identity testing:
  - Target binding methods alone may not be sufficiently specific for identity testing of an ADC produced in a facility where mAbs binding the same target are manufactured. A bridging ELISA may be used to capture both the payload and target specificity.
  - Alternatively, additional methods (for ADC-specific attributes) may be needed to ensure proper identity testing, such as the combination of target binding and payload detection, charge variants, conjugation-specific attributes, or DAR.
- New types of payloads:
  - Different methods may be needed and should be appropriate for the payload-linker (e.g., oligonucleotides).
- Serum stability:
  - An assessment of the ADC serum-stability is expected in Phase 1 INDs.

Case Study - Justification for Control of Effector Function

- ADC consisting of IgG1 mAb and cytotoxin:
  - Original BLA claimed that effector function is not relevant to the MOA, while in vitro data indicate FcγR binding and moderate ADCC activity. No ADCC control in specification.
  - First IR response: A specification test for ADCC is not needed because 1) low contribution of ADCC to overall MOA (in vitro cell-killing data of mAb and ADC showed the cell killing potency of mAb is about 10% of that of ADC), 2) presence of cell killing test in the specification, 3) consistent glycan profiles observed during past manufacturing
  - Second IR response: 1) In vitro data showing comparable ADCC activity between mAb and ADC, 2) in vivo data showing that the primary MOA of the ADC is targeted delivery of the cytotoxin (mAb including its ADCC activity contributes minimally to the MOA), 3) consistent glycan data from 24 at-scale GMP batches, 4) comparable glycan data from small-scale worst-case materials with NOR/PAR approaching their limits, 5) commitment to include glycan profile testing in analytical comparability for the mAb if post-approval changes are made to the manufacturing process.

Acknowledgement

- Nailing Zhang
- Yan Wang
- Ashutosh Rao
- Emily Jing


FDA
U.S. FOOD & DRUG
ADMINISTRATION