White Paper

# Addressing the Unique Challenges of Data Management in ADC Development

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# Introduction

Antibody Drug Conjugates (ADCs), or immunoconjugates, are an increasingly important sub-class of antibody-related therapeutics. ADCs leverage the specificity of monoclonal antibodies (mAbs) to enable targeted drug delivery to tumor cells while minimizing or eliminating damage to healthy tissue, thus reducing the disruptive side effects associated with traditional chemotherapy drugs.

In addition to their ability to directly target tumor cells with a cytotoxic drug, ADCs are also showing promise for their ability to indirectly treat tumors by delivering immunogenic molecules that can stimulate the innate immune response in the tumor microenvironment.

While the vast majority of ADCs in clinical development are focused on oncology, there is growing interest in their potential to treat other diseases, including metabolic disorders and infectious diseases, due to their ability to overcome some of the issues associated with traditional small molecule therapies such as clearance and cellular internalization.

There are currently nine cancer ADCs on the market, the majority of which have been approved in the past few years. Growing interest in the potential of ADCs is clear from the fact that at least 50 biopharmaceutical companies have ADC development programs, with more than 180 ADCs currently in clinical trials. A recent report also predicts the ADC market to grow from the current level of $2-3 billion to exceed $13 Billion by 2026.[1]

Despite the therapeutic potential of ADCs, they are not without their challenges, which mainly arise due to their molecular complexity. In this whitepaper we will explore the challenges associated with the management of the large volume and variety of analytical data generated throughout the ADC development process.

Let's begin by exploring the main components of an ADC, discussing some of the unique challenges and how they may be overcome.

# The Anatomy of an Antibody-Drug Conjugate

Although some ADCs involve a direct linkage between the antibody component and the drug, it is far more common for an ADC to consist of three main structural units as illustrated in Figure 1: a monoclonal antibody (mAb), a small molecule drug (often referred to as the 'payload') and a linker covalently connecting the mAb to the payload.

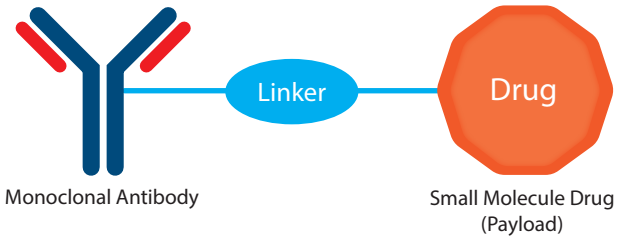


Figure 1. The three main components of an ADC: The mAb, the linker, and the 'payload'.[2]

The selected mAb must bind to antigens that are selectively expressed on the target, but the characteristics of the linker and drug components of the ADC are equally important.

There are two main routes to achieving the linkage:
i) Combine the linker and drug by chemical synthesis before conjugation to the mAb
ii) First couple the linker to the mAb then attach the drug to the linker

In either case, the linker is normally attached via exposed cysteine or lysine residues on the mAb.

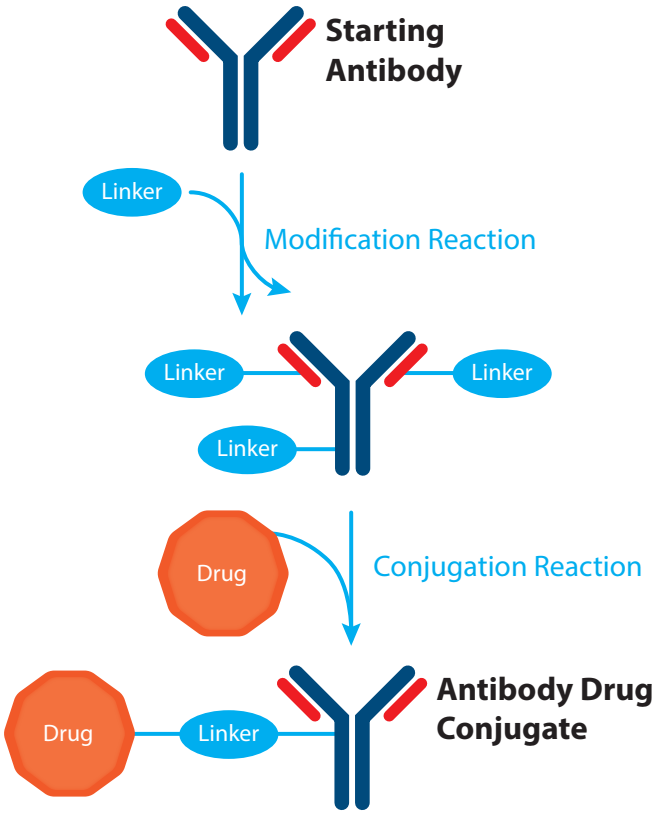


Figure 2. An example of one route to the production of an ADC[3] where the linker is attached to the mAb before attachment of the drug.

Maintaining a stable linkage between the mAb and drug during circulation in the blood is critically important since any free drug (or drug-containing degradation product) that is released from the ADC prematurely away from the intended site of action could increase the risk of non-specific toxicity. The conjugation process must also be predictable, resulting in a homogeneous product where a well-defined number of drug molecules are attached to the mAb, commonly referred to as the Drug Antibody Ratio (DAR).

The properties of the drug are also important to consider. Highly hydrophobic molecules have the potential to alter the biological properties of the mAb, which could result in aggregation of the antibody component either during the conjugation process or storage. Although the ADCs in the clinical development are directed against a relatively wide range of antigens, the difficulty in finding suitable drugs for ADCs is exemplified by the focus on a narrow range of drug classes such as auristatins, calicheamicins, duocarmycins, maytansinoids, and pyrolidobenzodiazepines.[4]

## Characterization of ADCs

Finding the optimal combination of ADC components to target a specific indication is a rather challenging balancing act. There is a strong focus on maximizing specificity, homogeneity, and stability whilst keeping immunogenicity to a minimum. Whichever specific combination of

components is selected, the structural complexity of the resulting molecule increases the need for comprehensive characterization of ADCs.

The goal of ADC development is to ensure a reproducible, consistent, scalable production process that yields a stable, high-quality product whilst minimizing chemistry manufacturing and control (CMC) complexity and optimizing drug-like properties.

To achieve this, it is important to understand the properties of each of the individual components of an ADC and the characteristics of each step of the production process; including attachment of the drug and linker, conjugation to the mAb, and subsequent purification steps, as well as the resulting process intermediates.

## Factors Affecting Efficacy

Many different factors determine the efficacy of the final product, including the homogeneity and stability of the final conjugated product. Homogeneity is typically expressed in terms of the average number of drug molecules attached to the mAb, otherwise known as the Drug to Antibody Ratio (DAR), and the distribution profile of the drug on the mAb, otherwise known as payload distribution. These aren't the only measurements of product homogeneity but they are particularly important as they also determine the amount of drug delivered to the target tissue(s), which directly impacts both efficacy and safety. Hence, they are not only important to determine during the development of an ADC, they are also critical measurements to monitor as part of the QC release process to ensure safety and minimize batch to batch variability.

The characterization of an ADC at each stage of its development cycle, from discovery through to preclinical and clinical development, is a complex undertaking which involves a range of complementary, orthogonal analytical techniques including UV/Vis spectroscopy, liquid chromatography, electrophoresis, and mass spectrometry. The specific blend of methods required is dependent on both the chemistry of the drug-linker and the method of conjugation. For example, Hydrophobic Interaction Chromatography (HIC) is often used to determine the DAR and drug loading distribution of cysteine-conjugated ADCs but is typically focused on molecules that have a limited number of potential conjugation sites.

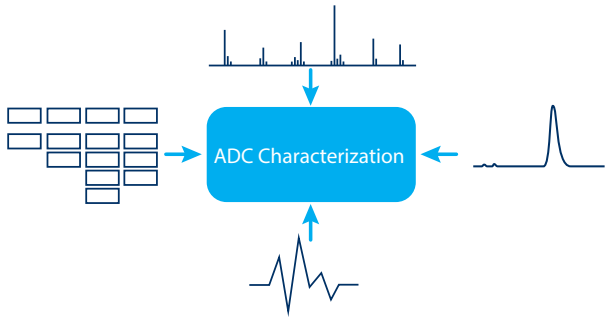


Figure 3. A variety of analytical techniques are used to characterize the structure of an ADC, the drug antibody ratio (DAR), and associated impurities.

## Understanding the Impurity Profile of ADCs

A range of different impurities, both mAb- or small molecule-related, can arise during any stage of the ADC production process. Small molecule impurities typically originate during manufacture of the drug, linker, or the linker-drug intermediate, such as residual solvents, reagents, and by-products, but can also form during manufacture or storage of the ADC drug substance or formulated drug product.

The typically cytotoxic nature of the ADC payload makes it critical to determine the levels of these impurities throughout the entire ADC manufacturing process, hence control strategies tend to focus on the minimization of small molecule impurities.[5] That's not to say, however, that large molecule impurities are not important. Degradation products may impair the ability of the mAb component to specifically bind to the target epitope and aggregates can affect the solubility of the product, both of which negatively impact efficacy of the ADC.

## The Challenge of Managing ADC Characterization Data

A common challenge for analytical groups is that instruments of the same analytical technique from different vendors generate data in different file formats and require their own dedicated software for data processing, extraction of results, and analysis. In some cases, different applications are used for the initial capture of raw data and its subsequent processing. Given the wide range of analytical techniques employed during the development and manufacture of a typical ADC, the associated analytical data is typically spread across multiple disconnected instrument databases, CDSs, and other data silos.

This problem is often compounded by the fact that in many biopharmaceutical companies the analytical methods focused on large molecule characterization are performed by different teams to those focused on small molecule analytics. Data is therefore further scattered within a project.

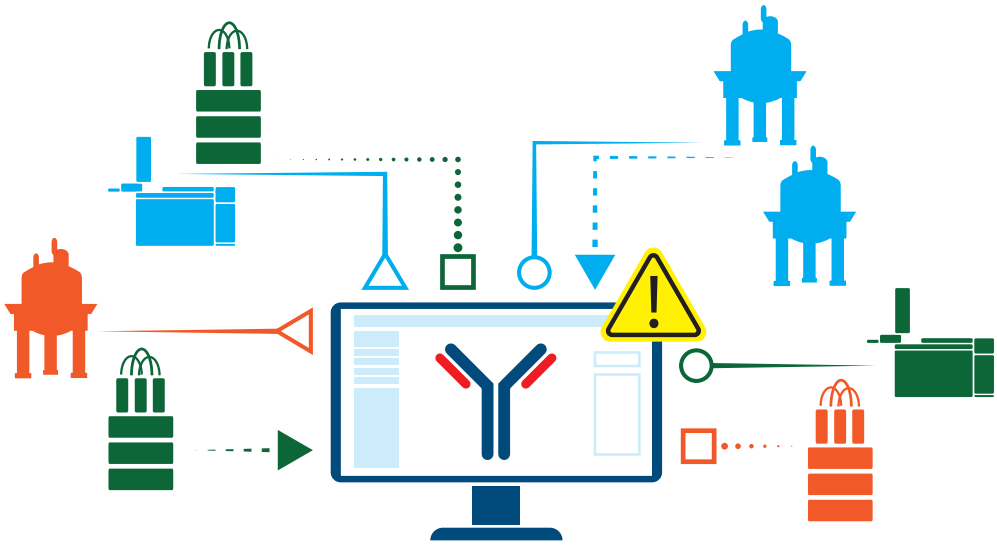

Figure 4. Data assembly from a variety of systems is a time-consuming and laborious task prone to errors.

This presents a significant challenge for organizations developing novel ADCs. A consequence of the volume and variety of analytical data, and the range of different data sources, is that bringing all relevant information together is a manual, time consuming, and error-prone activity that is sometimes only done at the point of reporting rather than as an ongoing process.

Some organizations have deployed Electronic Laboratory Notebooks (ELNs) to help centralize analytical data and improve its findability. However, this doesn't address the need for scientists to easily identify correlations between data generated by different analytical methods. Likewise, applications focused on managing data across different analytical methods and instruments typically focus on either small molecule or large molecule impurities, but rarely both.

The ability to characterize an ADC and understand changes to its impurity profile throughout the different stages of production and purification relies on the ability to accurately and explicitly link analytical data to specific contaminants and degradation products. Since these can be derived from the drug, linker, mAb, or various combinations of two or more of these components, visualization of their structure is an important consideration. While chemical structures are not an effective way to represent the mAb component, sequence-based representations cannot be used to represent the drug.

## The Role of Luminata in ADC Development

ACD/Labs understands the complexity of Antibody Drug Conjugate (ADC) development, especially the pains relating to effective data management. Luminata®, a software tool developed to support decision-making in process development, delivers a single integrated application to handle complete data management for ADC development

Luminata delivers a vendor-agnostic, multi-technique platform which enables scientists to manage, analyze, and visualize data from a wide range of analytical methods, and collaborate more effectively throughout the ADC development lifecycle. It provides a systematic and real-time view of all analytical data. It supports ADC development from the initial establishment of the most advantageous combination of ADC components through optimization of the conjugation and purification process, characterization of the final product and process intermediates, and understanding how process changes impact the impurity profile and efficacy of the product.

Luminata makes it easy to visualize and navigate the analytical data corresponding to each stage of the ADC production process. While enabling scientists to associate chemical structures for small molecules and related impurities with analytical data, Luminata also leverages HELM ( Hierarchical Editing Language for Macromolecules)[6] to provide scientists with an easy to understand representation of the ADC from which they can rapidly navigate from a high level classical 'Y-shaped' representation of the antibody to a detailed chemical structure.

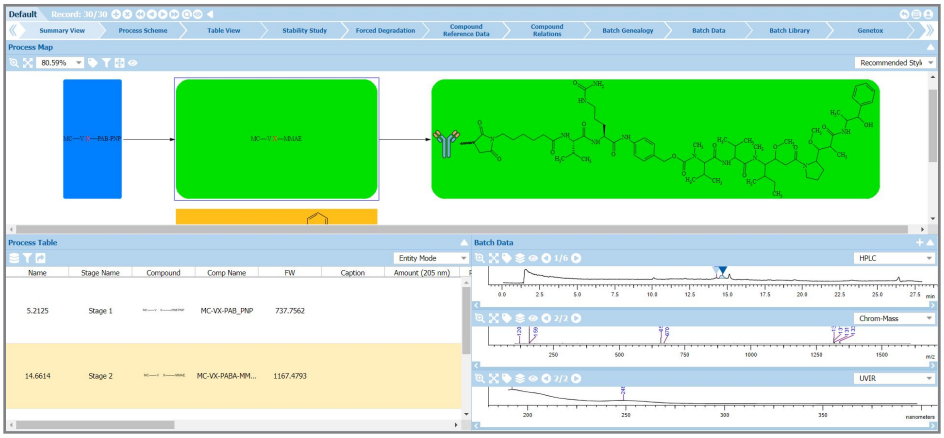


Figure 5. Luminata can be used to collect and connect all relevant drug, linker, and mAb information with relevant analytical data for centralized management of process development data.

Luminata enables scientists involved in the development and manufacturing of ADCs to see the big picture and make better informed decisions faster from a single software interface.

## References


1. Kuick Research. (**2019**, June 19). *Global Cancer Antibody Drug Conjugate Market, Price, Dosage & Clinical Trials Insight 2026*. https://www.medgadget.com/2020/06/global-cancer-antibody-drug-conjugate-market-price-dosage-clinical-trials-insight-2026.html

2. Adapted from Hutchison, M.H., Hendricks, R.S., et al. (**2018**). Chapter 40: Process Development and Manufacturing of Antibody-Drug Conjugates. In Jagschies, G, Lindskog, E, et al. (Eds), *Biopharmaceutical Processing: Development, Design, and Implementation of Manufacturing Processes* (pp. 813–836). Elsevier. DOI: https://doi.org/10.1016/B978-0-08-100623-8.00041-4

3. Adapted from Rohrer, Thomas K. (**2013**, June 21). *Consideration for the Safe and Effective Manufacturing of Antibody Drug Conjugates*. ADC Review. https://www.adcreview.com/articles/consideration-safe-effective-manufacturing-antibody-drug-conjugates/

4. Alain Beck & Janice M. Reichert. (**2014**). *Antibody-drug conjugates*. mAbs, 6(1): 15-17. DOI: 10.4161/mabs.27436

5. For example, see: Gong, H.H., Ihle, N., Jones, M.T., et al. (**2018**). *Control Strategy for Small Molecule Impurities in Antibody-Drug Conjugates*. AAPS PharmSciTech., 19(3): 971–977. doi:10.1208/s12249-017-0943-6

6. *HELM – Pistoia Alliance*. (n.d.). Pistoia Alliance. Retrieved October 14, **2020**, from https://www.pistoiaalliance.org/projects/current-projects/helm/


## About the Authors


Dr. Eliot Randle is an independent consultant in the Biopharmaceutical industry. He has over 25 years industry experience, and for the last 15 years has focused on helping companies transform their operations through better use of informatics. Eliot has held several senior roles within technology companies and holds a B.Sc. in Biochemistry, a Ph.D. in Molecular Biology from the University of Manchester, UK, and an MBA from Warwick Business School, UK.



Joe DiMartino is the Luminata Solution Manager at ACD/Labs. Prior to his current role, he was part of the Technical and Scientific Support Team, where he worked as a Senior Application Scientist.

Previous to his career at ACD/Labs, Joe worked in drug development at Apotex Pharmachem, Inc. His responsibilities included the design of optimized procedures for the synthesis of active pharmaceutical drugs. He was also an integral part of a team that synthesized potential impurities. Joe earned his Bachelor of Science at the University of Winnipeg, and his Graduate Degree in Chemistry at the University of Windsor.