Antibody-Drug Conjugate Research and Development: The Role of Multifaceted Mass Spectrometry
(ASMS 2017. Indianapolis, IN. Pharmaceuticals Interest Group)

John Valliere-Douglass (Seattle Genetics) and Matt Schenauer (Gilead Sciences) presided over the workshop which took place on the evening of Monday, June 5th.

The workshop was well-attended with few free seats in the combined, 141-142, conference rooms. An expert panel comprised of 8 individuals along with the moderators participated in a robust discussion on various topics surrounding the applications of MS and analytical approaches in ADC R&D.
Following a brief initial introduction to the field and brief touchpoints of where/how MS analysis fits into the ADC R&D landscape, discussion ensued.
Members of the audience ranged in levels of exposure to the field, from novice to seasoned expert. Sample question/discussion points were also drawn from audience interest based on a preconference survey, which served to bridge brief gaps in between live audience questions. Overall topics ranged from approaches/applications of MS in structural characterization to regulatory approaches on integrating MS data in quality control environments normally dominated by immunochemical, chromatographic, spectrophotometric, and electrophoretic techniques. Questions related to applications of newer MS-based approaches (e.g. CE-MS, HDX) and highly complex areas of traditional method/MS method harmonization (e.g. host cell protein analysis) were also raised and discussed.
This year the expert panels consisted of the following contributors: Cong Wei, Vertex Pharmaceuticals; Andrew Dawdy, Pfizer; Shawna Hengel, Seattle Genetics; Zhiqi Hao, Genentech; Omar Hamdy, Abbvie-Stemcentrx; Christina Malinao, Agensys; Jintang He, Genentech; and Jason Hogan, Bristol-Myers Squibb. The co-chairs of the sessions would like to thank the panelists and the audience for a lively discussion. We hope you found it as beneficial and as informative as we did!

A basic theme that emerged from the workshop was the importance of understanding the stability of the drug-linker, mAb and ADC in various matrices. It was stressed that when undertaking characterization exercises, method developers should assess the impact of the method conditions on the molecule and choose conditions that minimize artifactual (method induced) degradations. Specifically, method developers should understand the extent to which rp-HPLC separations carried out under extremes of temperature and pH might degrade the drug moiety of the ADC during detection/characterization.

On reconciliation of MS and ELISA based methods for characterization and quantitation of host-cell proteins: there was some discussion on this topic and the point was made that, a priori, there is no reason that the MS based result should (quantitatively) align with the ELISA result. This is because MS detection is (in theory) unbiased while ELISA detection and quantitation is based on immuno-reactivity of the HCP(s).

On the topic of detection and quantitation of amino acid sequence variants in mAbs by MS; there was broad consensus on the overall approach which was to analyze samples by peptide map and perform data searches using an error tolerant type algorithm capable of detecting changes in primary sequence that are consistent with amino acid substitutions. There was not a general consensus on particular software suites to use for this work although various vendor-specific and 3rd party software packages were mentioned. Emphasis was placed on the need to have software capable of identifying and quantitating sequence variants and presenting the output in a manner that facilitated rapid screening of samples.

Native mass spectrometry techniques for the determination of the intact mass of ADCs continue to be a hot topic. Most practitioners are using online SEC to exchange the ADC from its formulation buffer into a volatile salt buffer that is mass spec friendly. On the choice of using native methodologies, participants indicated that the approach was called for when analyzing non-covalent (interchain cysteine linked) ADCs and ADCs employing a drug linker subject to poor recovery and/or on-column degradation during analysis. Some participants indicated that native MS techniques could be qualified for the determination of ADC drug to antibody ratio (DAR) but that this might not be straightforward if there was not an obvious orthogonal methodology to benchmark against.

There was some discussion about the use of MS based higher order structure (HOS) methodologies such as HDX and amino acid footprinting strategies. While there was no indication that these methodologies are being used routinely in development and/or filings, it was indicated that having this data for particular classes of modalities could be useful for demonstrating that drug conjugation did not have an impact on the overall HOS of the parent mAb.