

*ADC Research and Development: The Role of Mass Spectrometry in ADC Biotherapeutic Development
(ASMS 2018. San Diego, CA. Pharmaceuticals Interest Group)*

John Valliere-Douglass (Seattle Genetics) and Andrew Dawdy (Pfizer) presided over the workshop which took place on the evening of Wednesday, June 6th.

Abstract

ASMS workshop: ADC Research and Development: The Role of Mass Spectrometry in ADC Biotherapeutic Development (Pharmaceuticals Interest Group)

Recognizing the recent significant clinical and commercial success of ADCs, the pharmaceutical interest group initiated this workshop to explore the role of mass spectrometry in ADC R&D. The format of the workshop will consist of a short informal presentation (less than 15 minutes) followed by an audience driven discussion with peers and a panel of experts. The short presentation will include a primer on ADCs, for those practicing MS but unfamiliar with ADC therapeutics and their conjugates, and then provide a snapshot of current applications of MS analysis in the industry for ADC R&D. The organizers will have backup questions prepared for the panel and audience to start or prompt the discussion if needed. Potential areas of discussion may include initial mAb and drug assessments, bioanalytical assay development and the scaling range of characterization required for ADCs as they progress through clinical development. Discussion may focus on MS method development, optimization, data analysis, and how this information is being applied within industry paradigms or changing them.

We estimate the workshop, taking place in room 33 ABC, had about 75 attendees. An expert panel comprised of 6 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. The workshop began with a brief primer on ADCs, including a comparison/contrast of the four commercially approved ADCs in terms of their clinical targets, conjugation type, and payload class.

The audience contained some seasoned experts, but a show of hands revealed that only about a quarter of the attendees were working with ADCs themselves. Sample questions and discussion points were drawn from audience interest based on a preconference survey as well as from the facilitators and panelists prior to the workshop. Overall topics ranged from approaches/applications of MS for heightened characterization, determination of DAR, and comparison of MS quantitation to that of LBAs, to challenges associated with ADC stability. This year the expert panels consisted of the following contributors: Chris Chumsae, Abbvie; Alice Chin, Seattle Genetics; Linjie Han, Abbvie; Michael Kim, Genentech; Lisa Marzilli, Pfizer; and Lintao Wang, Immunogen. The co-chairs of the sessions thank the panelists and the audience for their questions and sharing their knowledge and experience as it relates to ADC development.

The pre-conference survey indicated that one of the top challenges encountered by those working in ADC research and development was in vitro, in vivo, and in formulation stability. The importance and challenge of properly designing relevant forced degradation studies and the appropriate use of surrogates in stability studies was discussed. Also when it comes to characterization, method developers should assess the impact of the method conditions on the molecule and choose conditions that minimize artefactual (method induced) degradations. Appropriate conditions may be slightly or

dramatically different than those in platform methods used for modalities such as mAbs. An additional method development challenge touched on during the workshop was the hydrophobicity of linker-payloads.

Another voiced challenge related to stability is the quantitation of small molecules such as drug-related impurities. The use of MS in quantitation was discussed for some time, including the comparison of MS to ligand binding assays for quantitation. Measuring free-drug, free mAb, and conjugated mAbs was discussed as well as selection of appropriate internal standards to quantitative assays.

Gaining 100% or appropriate sequence coverage was another top challenge reported from the pre-conference survey. However, there was not much discussion on this topic during the workshop itself. Sequence variant analyses were not discussed this year either.

The use of native-ESI mass spectrometry continues to show major growth based on the survey and workshop discussion. Situations for the use of native vs denaturing intact mass analysis were discussed. 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. Also native or lesser acidic (formic- vs TFA-based) forms of denaturing mode can be used when characterizing labile linker-payloads that are subject to in-source fragmentation or on-column degradation. Due to the increased quality of intact MS and software aids in recent times, some are quantitating DAR using intact MS, but ionization of different DAR populations and how well MS compares to other methods like HIC was still brought up as a concern. Sample heterogeneity and the impact of peak broadening and mass shifts at the intact level were also discussed.

Questions regarding the use of top-/middle-down techniques in relation to ADC characterization were raised during the workshop. It appeared that these are not widely implemented yet for ADCs.

A suggestion from one of our panelists is to encourage better introductions of not only the panelists but also participating audience members in the future. The intention of this would be to foster a more collaborative environment during the session rather than a panelist vs audience member mentality. Overall, the workshop fostered good discussion and was a success.

Based on the workshop demographics, survey feedback, and the current landscape of biopharma, we may give consideration to expanding the focus of the future workshop beyond ADCs to other types of bioconjugates or mAb-like modalities such as bispecifics in order to address the increasing breadth of modalities in biopharma. We will aim to best capture what is “hot” and of current interest within the Pharmaceutical Interest Group.