Saturday and Sunday (Two days)

Antibody-Drug Conjugates, Oligonucleotides, Peptides, and Other Complex Drug Modalities Characterization and Quantification by Mass Spectrometry

Instructors: **Ragu Ramanathan** (Pharmaron US Labs and Cell & Gene Therapy, Exton, PA) and **Anton Rosenbaum** (AstraZeneca, South San Franciso, CA)

Small-molecule drugs have dominated the disease treatment landscape over the past several decades. However, to treat complex diseases, biopharmaceutical companies have developed and received approvals for the use of an increasing number of complex drug modalities, including antibody-drug conjugates (ADCs), oligonucleotides (ONGs including ASO, SiRNA, mRNA, plasmid DNA, etc.), peptides, and biologics, which have been approved over the past 25 years. Of the pharmaceutical drugs approved from 2015-2024, the complex drug modalities represent 20-40% of the portfolio. Mass spectrometry-based strategies are extensively used to quantify and characterize ADCs, ONGs, peptides, and biologics. This course is designed as an introduction for biopharmaceutical researchers, academicians, and regulators needing to expand their knowledge of using mass spectrometry-based methods to quantify and characterize multiple end points of complex drug modalities.

The overall complex drug modality drug discovery and development process will be divided into seven stages to introduce LC-MS-based quantification and characterization studies required at each stage: (1) hit to lead, (2) lead optimization, (3) candidate selection, (4) preclinical development, (5) clinical development, (6) registration and launch (7) post-approval. The key bioanalysis and DMPK-related activities at each stage will be discussed, and the similarities and differences will be highlighted compared to those of small molecule drug discovery and development.

Current triple quadrupole (QqQ) and high-resolution mass spectrometry (HRMS) instrument options will be described and compared, including quadrupole-time-of-flight (Q-TOF) and Fourier transform mass spectrometry (FTMS). Mass analyzer options will be extended to MS/MS platforms, including some hybrid instruments. Qualitative and quantitative applications of HRMS will be discussed, especially for ADCs, ONGs, peptides, and proteins. These applications will include molecular formulae and structure analyses, such as identifying metabolites, posttranslational modifications, and the effects of high resolution on sensitivity and specificity in quantitation.

In addition to LC-MS, ligand-binding assays (LBA) using meso scale discovery (MSD) and enzyme-linked immunosorbent assay (ELISA), along with cell-based assays, used to evaluate the pharmacokinetic (PK) and immunogenicity properties of complex drug modalities will be discussed. The roles of flow cytometry and real-time quantitative polymerase chain reaction (Q-PCR) assays will be discussed in the context of bioanalysis. Case studies will illustrate strategies for measuring some bioanalytical endpoints using LC-MS and LC-HRMS techniques instead of LBA or PCR-based assays. By the end of the course, you'll be ready to apply your knowledge in real-world scenarios and contribute to interdisciplinary efforts to solve complex biomedical problems.

Prerequisite: Hands-on experience in mass spectrometry or related bioanalytical techniques or completing the ASMS online short course "High-Resolution Mass Spectrometry for Qualitative and Quantitative Analysis: An Introduction."