

Table 5.1 Biopharmaceutical Tools Peculiarities and Attributes

Technique	Peculiarity	Attributes
MS		
Hydrogen deuterium exchange MS	Global and local amino acid level structure and peptide backbone	Changes in HOS; dynamic structure
Charge state distribution	Gas-phase global structure	Changes in HOS
Covalent labeling (footprinting)	Global and local amino acid level structure with side chain structure	Changes in HOS; dynamic structure
Ion-mobility spectrometry	Gas-phase global structure	Changes in HOS
Spectroscopy		
UV	Aromatic amino acid environment	Biophysical and HOS; the FDA requires calculation of extinction coefficient
CD	Peptide bond structure and aromatic amino acid environment	Biophysical and HOS; near- and far-UV spectroscopies provide different, secondary and tertiary attributes
Fluorescence spectroscopy	Aromatic amino acid environment; most proteins have the three fluorescing amino acids	Biophysical and HOS; also, ideal test for stressed solution evaluation for fourth-dimensional evaluation; fluorescence microscopy is an emerging field to evaluate
Fourier transform infrared (FTIR) spectroscopy	Peptide bond structure	Biophysical and HOS
NMR	Atomic level resolution via ^1H , ^{13}C , ^{15}N NMR	Changes in HOS; dynamic structure evaluation
Chromatography		
IEXC	Surface polar and charge interactions	Biophysical and HOS
Hydrophobic interaction chromatography (HIC)	Surface nonpolar interactions	Biophysical and HOS
Simple interval calculation	Surface interactions with itself	Biophysical and HOS
Affinity	Surface interactions with binding partner, ligand	Biophysical and HOS
Electrophoresis		
Native gel	Global net charge and structure	Biophysical and HOS; aggregation
Native IEF (imaged capillary IEF)	Global net charge	Biophysical and HOS
Native CZE	Global net charge and structure	Biophysical and HOS; aggregation
SDS-gel/capillary	Size, covalent aggregation	Biophysical and HOS; aggregation
Thermodynamics		
DCS	Global and domain structure	Biophysical and HOS
Isothermal titration calorimetry	Protein binding	Biophysical and HOS

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