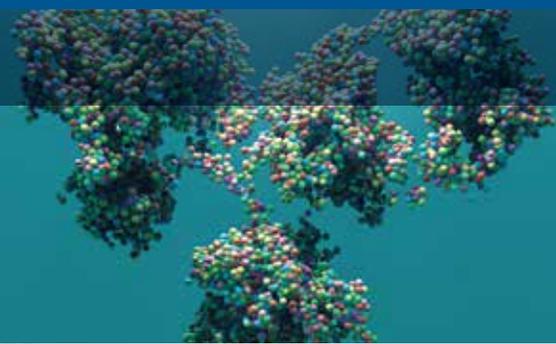


Enabling Routine Characterization of Proteins

Agilent MassHunter BioConfirm software



BioConfirm—a Key Component of Your Major Biopharma Workflows

Intact protein analysis

BioConfirm will rapidly deconvolute your intact protein data to determine molecular weights and confirm post-translational modifications (PTMs). PTMs like glycoforms can be labeled on the mass spectrum and inspected by their relative quantitative amounts using tables or histogram plots (Figure 1). Additionally, Cysteine disulfide bonds can either be individually specified in the protein sequence or just the total number listed—speeding up your setup time!

Sample prep:
AssayMAP Bravo

Analyze:
BioConfirm Software



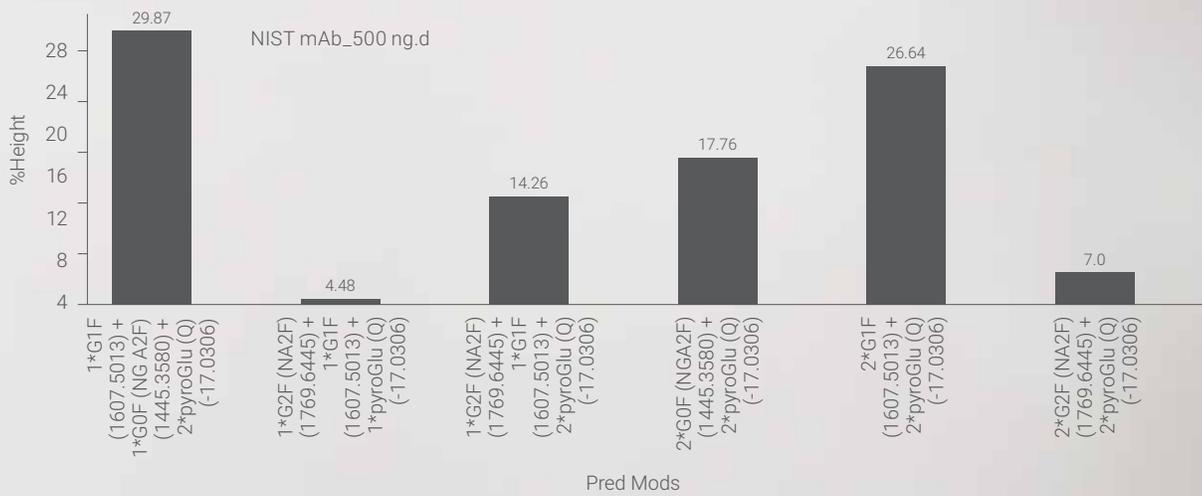


Figure 1. Relative quantitative results of glycoforms are displayed as histogram plots, allowing you to monitor a process.

Separation:
1290 Infinity II UHPLC and
AdvanceBio columns

Detect:
6545XT AdvanceBio LC/Q-TOF

Peptide Mapping

With MassHunter BioConfirm you can perform sequence confirmation with confidence. The peptide mapping algorithm uses MS/MS data to narrow down the list of possible peptides—saving you time from manual inspection. Results are displayed in the Sequence Coverage Map that allows you to review multiple samples (e.g. different digest enzymes) at the same time (Figure 2). In addition, histogram plots enable the comparison of multiple PTMs or conditions. Fast disulfide bond mapping can be used to untangle the scrambling of disulfide bonds in a monoclonal antibody.

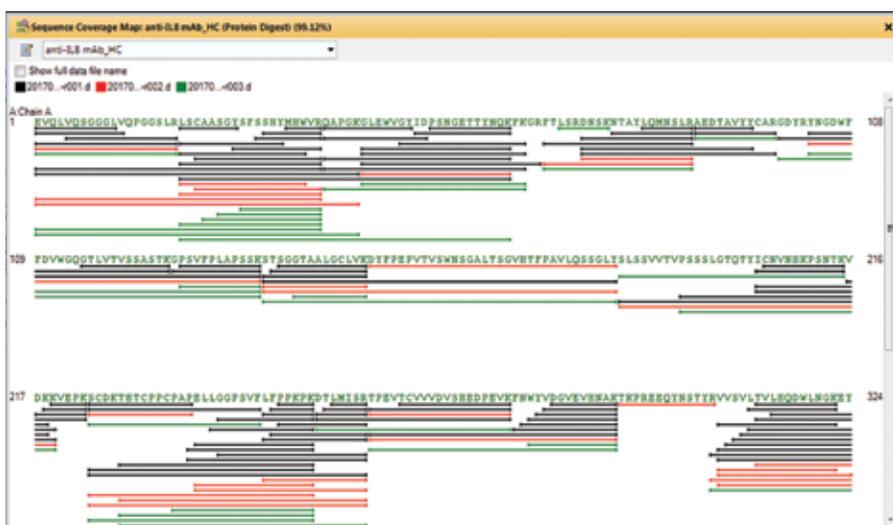


Figure 2. The Sequence Coverage Map makes review of multiple samples easy and clearly denotes MS/MS and MS-only evidence of the sequence.

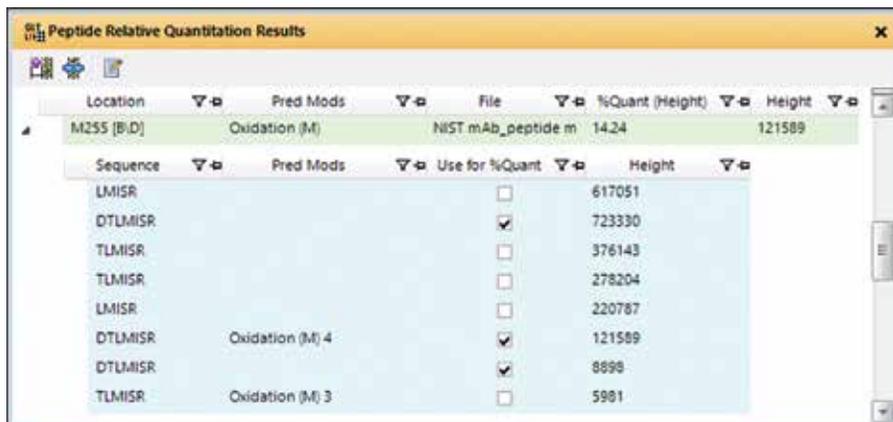


Figure 3. MassHunter BioConfirm allows you to do relative quantitation of modified and unmodified peptides.

Released Glycan Profiling

Glycans are a challenging PTM to characterize given the diversity in their composition and the need to chemically tag them to get adequate analytical sensitivity. BioConfirm has a Released Glycans workflow that allows for easy setup of the analysis and accommodates many commercial and custom tags. A curated database of glycans is provided that can be easily extended with custom glycans using MassHunter tools.

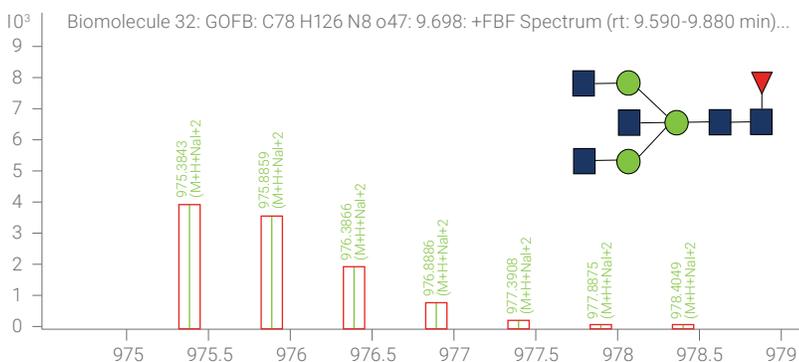
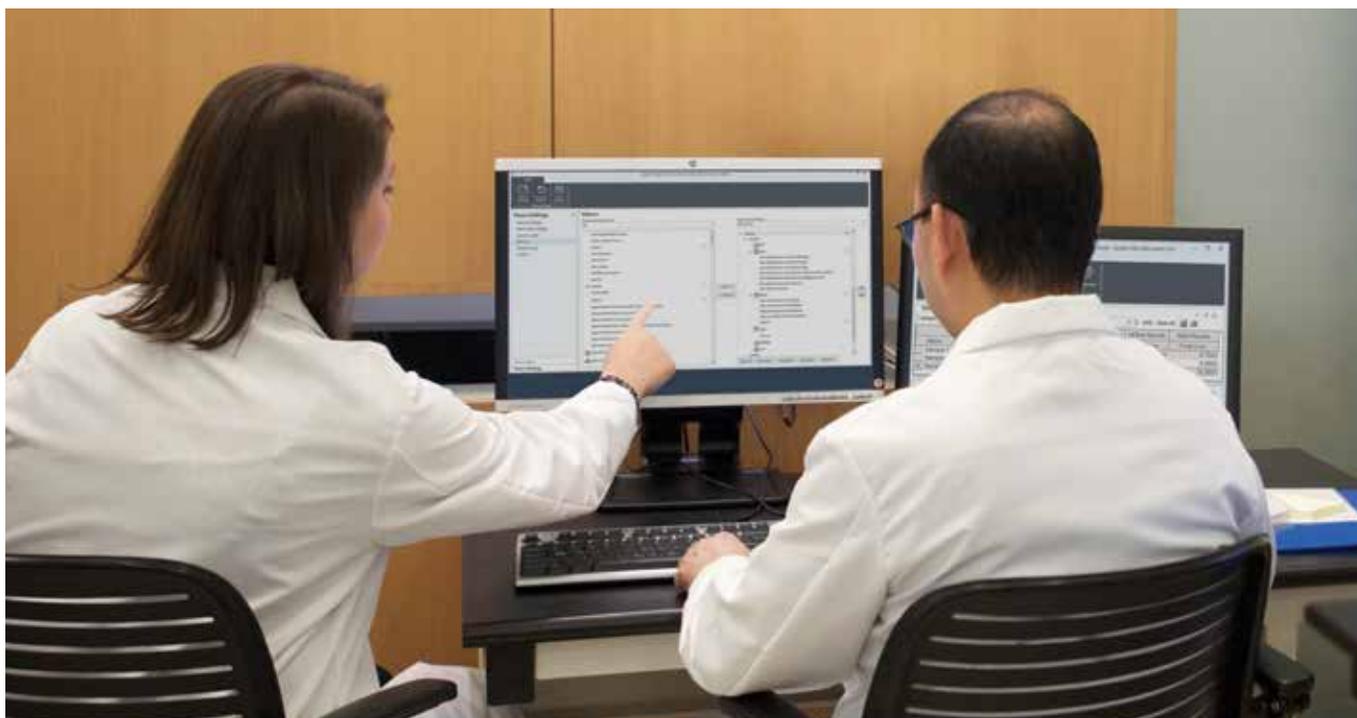


Figure 4. The released glycans results include spectra annotated with the glycan structure and theoretical isotope distribution.

One Click ADC DAR Calculation

The drug-to-antibody ratio (DAR) is a critical quality attribute of antibody drug conjugates (ADCs) that must be carefully monitored during the development process. MassHunter DAR Calculator has been created to rapidly calculate DAR values for both intact and reduced ADCs with minimal user input.



ADC Information

File Path	000001.D	DAR	11
Processing Method	Auto	Report Date	10/10/2018 12:18
Property	Theoretical DAR	Observed DAR	
Chemical Name	10000	10000	
Empirical Mass	900	900	

Deconvoluted Spectrum

DAR Peak	Peak	Theoretical Mass	Mass	% Area
1	10000	900	900	1.0
2	10000	1000	1000	1.0
3	10000	1100	1100	1.0
4	10000	1200	1200	1.0
5	10000	1300	1300	1.0
6	10000	1400	1400	1.0
7	10000	1500	1500	1.0
8	10000	1600	1600	1.0

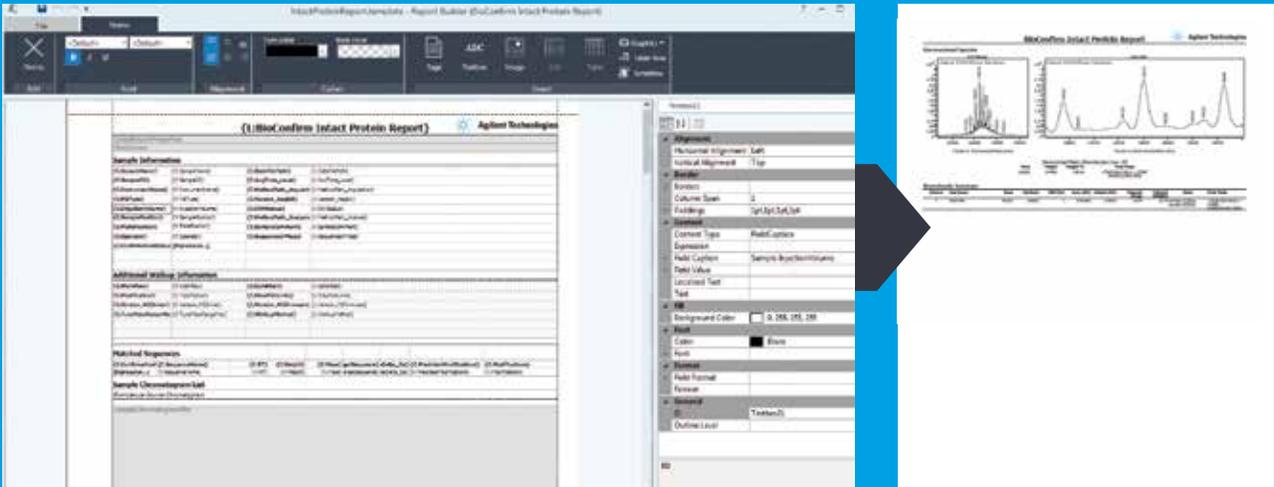
- Automatic calculation of DAR value
- Intuitive peak integration
- Built-in support for intact and reduced ADCs
- Dedicated ADC DAR report in PDF and MS Word

Walkup Software

The MassHunter Walkup environment provides sample submitters access to expert LC/MS capabilities without requiring them to be LC/MS experts. Biologists and Protein Scientists can automatically receive an annotated report with results for intact protein analysis, PTMs, peptide mapping and DAR values while having complete control of the sample's preparation, treatment and storage prior to analysis.

Reports Easily Done Your Way

The PDF Report Builder enables you to create reports in your preferred layout. Templates for all workflows are provided and can be modified to highlight the information that is important to you and your laboratory.



- Add your company logo
- Place tables, chromatograms, and spectra exactly where you want them
- Fast creation of reports in PDF format

Speeding Up Your Biopharmaceutical Analyses

MassHunter BioConfirm is an integral part of Agilent's biopharma workflows that span from sample preparation to separation and detection through data analysis and reporting. With support for intact protein analysis including glycoforms, peptide mapping using high confidence MS/MS evidence, and the released glycan capabilities, BioConfirm makes characterization of protein therapeutics fast and routine.

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