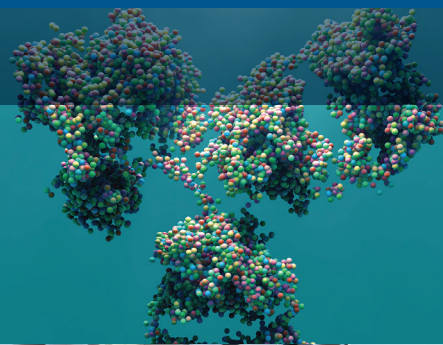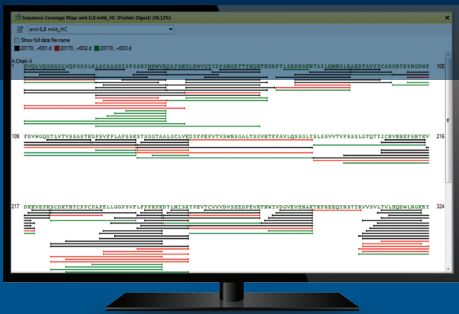# Protein Characterization Made Routine and Secure

Agilent MassHunter BioConfirm software

BioConfirm—Confidently Analyze for Major Critical Quality Attributes

Intact protein analysis

BioConfirm rapidly deconvolutes 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). Also, cysteine disulfide bonds can be either individually specified in the protein sequence or listed in total, speeding up your setup time.

Sample prep:
AssayMAP Bravo

Analyze:
BioConfirm Software

NIST mAb_500 ng.d

%Height (y-axis): 4, 8, 12, 16, 20, 24, 28

Predicted Modifications (x-axis)

Bar values and labels:
- 29.87 — 1*G1F (1607.5013) + 1*G0F (NG A2F) (1445.3580) + 2*pyroGlu (Q) (-17.0306)
- 4.48 — 1*G2F (NA2F) (1769.6445) + 1*G1F (1607.5013) + 1*pyroGlu (Q) (-17.0306)
- 14.26 — 1*G2F (NA2F) (1769.6445) + 1*G1F (1607.5013) + 2*pyroGlu (Q) (-17.0306)
- 17.76 — 2*G0F (NGA2F) (1445.3580) + 2*pyroGlu (Q) (-17.0306)
- 26.64 — 2*G1F (1607.5013) + 2*pyroGlu (Q) (-17.0306)
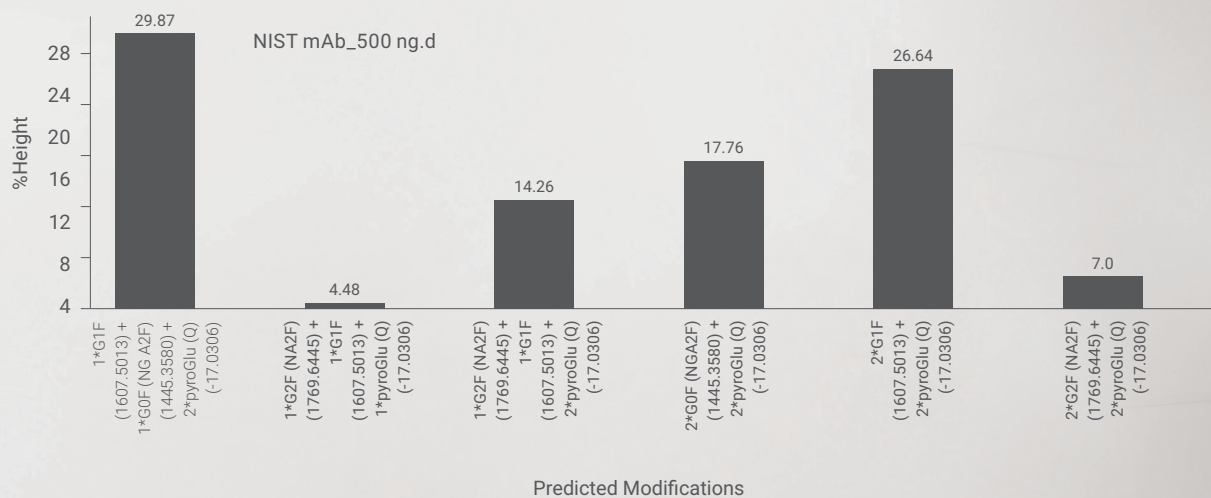- 7.0 — 2*G2F (NA2F) (1769.6445) + 2*pyroGlu (Q) (-17.0306)

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

Separation:
1290 Infinity II Bio LC and AdvanceBio columns

Detect:
6545XT AdvanceBio LC/Q-TOF

Peptide Mapping:
Confirm Sequences with Confidence

The MassHunter BioConfirm peptide-mapping algorithm uses MS/MS data to narrow the list of possible peptides—saving you manual inspection time. Results are displayed in a sequence coverage map that allows you to review multiple samples (such as different digest enzymes) at the same time (Figure 2). Histogram plots let you compare multiple PTMs or conditions. In addition, fast disulfide bond mapping can be used to untangle the scrambling of disulfide bonds in a monoclonal antibody.
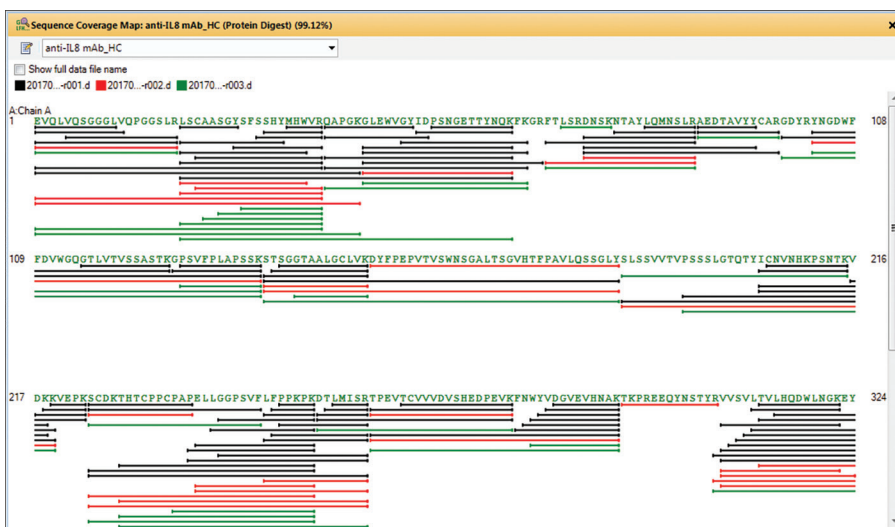
Figure 2. The sequence coverage map makes reviewing multiple samples easy and clearly denotes MS/MS and MS-only evidence of the sequence.
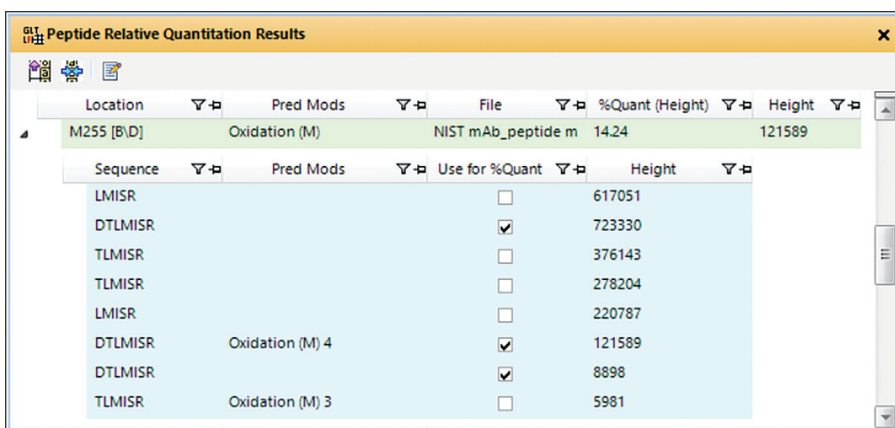

Figure 3. MassHunter BioConfirm allows you to do relative quantitation of modified and unmodified peptides.

Improve Sensitivity for Released Glycan Profiling

Glycans are a challenging PTM to characterize given the diversity in their composition and the need for chemical tagging to ensure analytical sensitivity. The BioConfirm-released glycans workflow makes setup easy and accommodates many commercial and custom tags. You can also take advantage of a curated glycan database, which can easily be extended with custom glycans using MassHunter tools. Learn more at **www.agilent.com/chem/glycan-sampleprep**
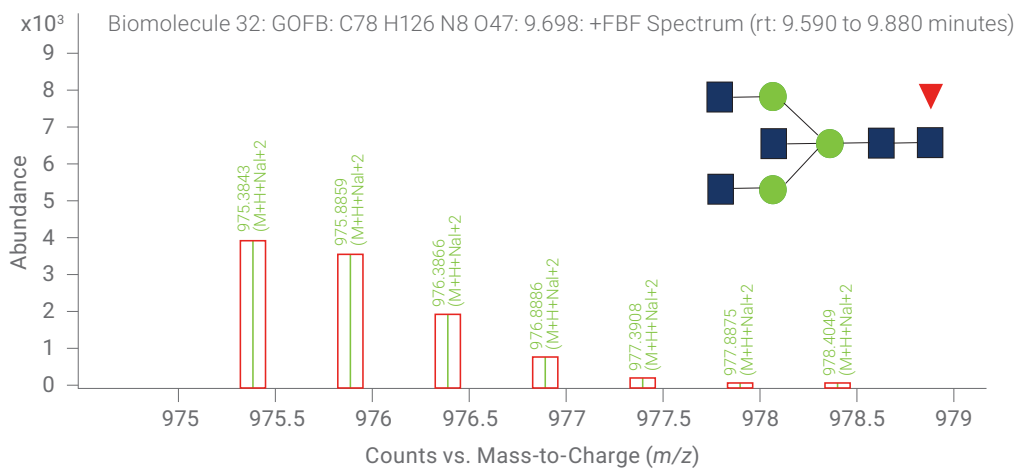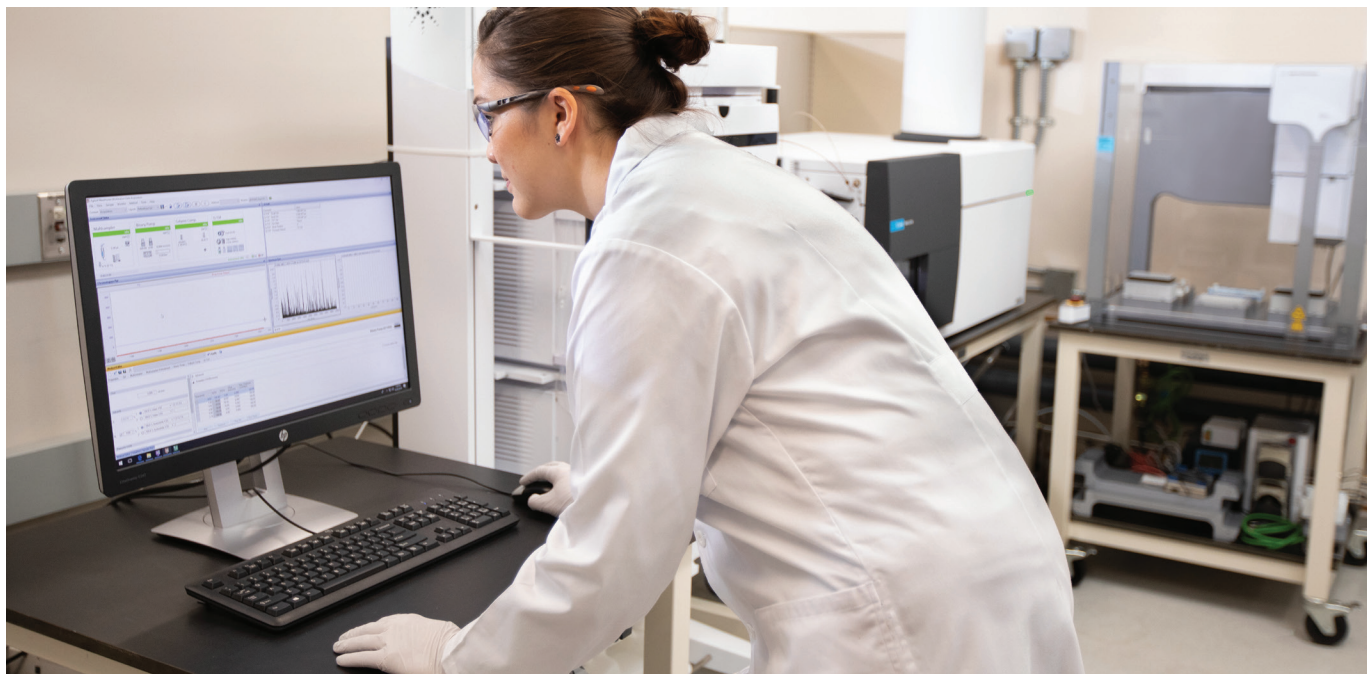

Figure 4. The released glycans results include spectra annotated with the glycan structure and theoretical isotope distribution.

Compliant-ready Features Keep You in Control

With advanced security features and built-in technical controls, BioConfirm Networked Workstation minimizes security risks while helping to preserve data integrity.

The new Control Panel allows you to set up roles and permissions to comply with regulatory guidance, such as 21 CFR Part 11 and Annex 11.
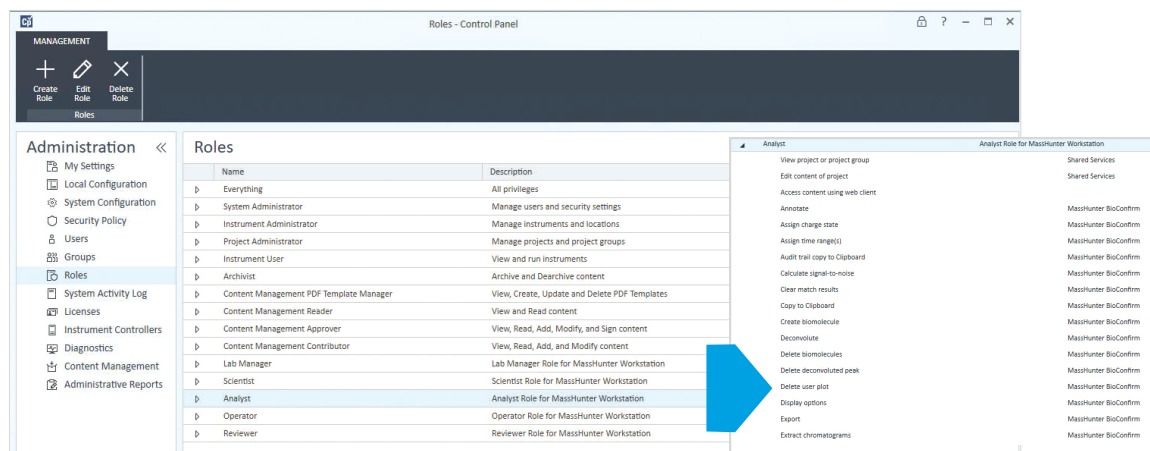

The Audit Trail makes reviewing easy by highlighting entries that require review. Tamper detection is automatically included using checksums.

Reports Done Easily—Your Way

The PDF report builder makes it simple 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.
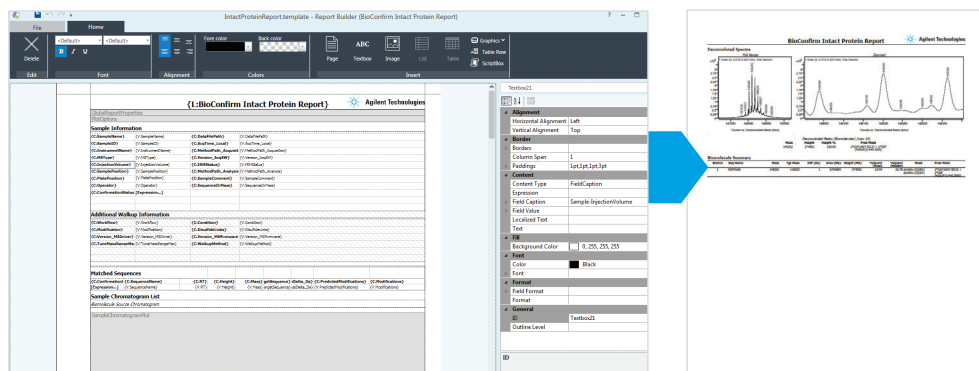
- Add your company logo
- Place tables, chromatograms, and spectra exactly where you want them
- Create PDF reports quickly

Speed Up Your Biopharmaceutical Analyses

MassHunter BioConfirm is part of the Agilent biopharma workflow, spanning from sample preparation, to separation and detection, through data analysis and reporting. This chart will help you choose the version of MassHunter BioConfirm that is best for your lab.

	Workstation	Networked Workstation
Software and license	•	•
Intact protein workflow	•	•
Protein digest workflow	•	•
Released glycans workflow	•	•
Recommended for GxP labs		•
Audit trails		•
Revisions		•
Access control		•
Server-based content management		•
Single-point access to data from multiple sources		•

Agilent CrossLab: Supporting Your Success

CrossLab is an Agilent capability that integrates services and consumables to support workflow success, improve productivity, and enhance operational efficiency. Through CrossLab, Agilent strives to provide insight in every interaction to help you optimize the return you get on your instrument investment and achieve your business goals. Agilent CrossLab supports Agilent instruments and select non Agilent instruments as well. We also provide consultative support for workflow enablement, lab analytics, regulatory compliance, inventory management, and asset management, including relocation services. Learn more about CrossLab at **www.agilent.com/crosslab**

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