

Confidently Analyze for Major Critical Quality Attributes (CQAs)

Understanding the attributes of a biologic drug, and the processes used to create it, is critical. [Agilent MassHunter BioConfirm software](#) is a biopharmaceutical software that enables routine characterization of complex biomolecules through easy-to-use workflows for oligonucleotide analysis, intact protein analysis, peptide mapping, and released glycan profiling.

Agilent MassHunter BioConfirm software, part of the complete Agilent biopharma workflow

Automate sample preparation

AssayMAP Bravo



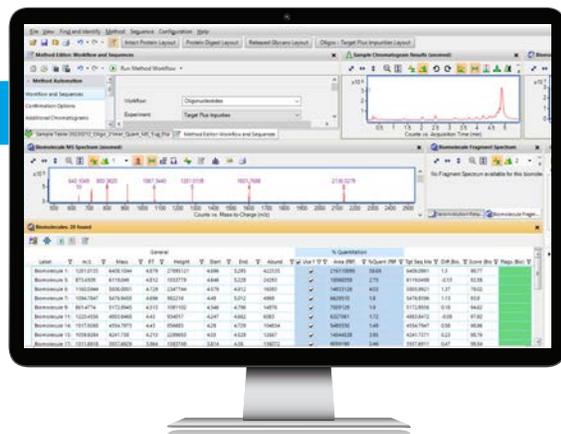
Separate

1290 Infinity II LC



Analyze and report

MassHunter BioConfirm software

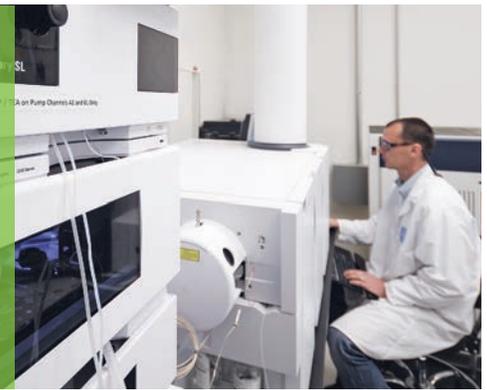


Detect

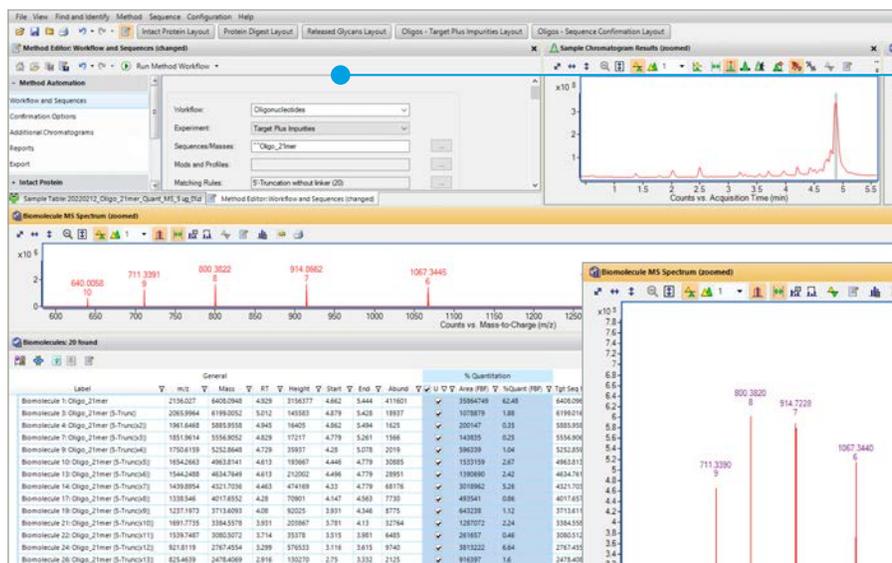
6545XT AdvanceBio LC/Q-TOF



Oligonucleotide Characterization: Elevate Your Expectations for Speed and Accuracy



BioConfirm software enables you to rapidly set up two workflows, Target Plus Impurities, and sequence confirmation for oligonucleotide confirmation.

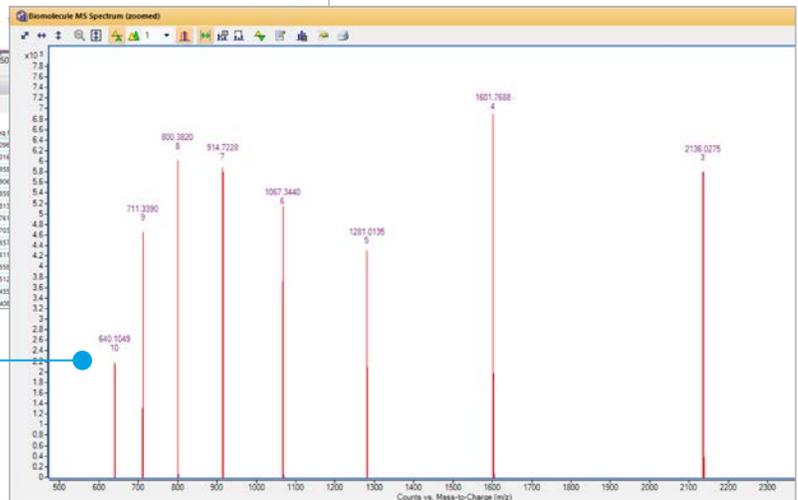


Full layouts

Full layouts have been added for oligonucleotides to rapidly and easily review results in chromatograms, spectra, and tables.

MS spectra

MS spectra show the oligonucleotide biomolecule annotated at each charge state.



Chemical Data Dictionary Editor

Proteins | Oligonucleotides

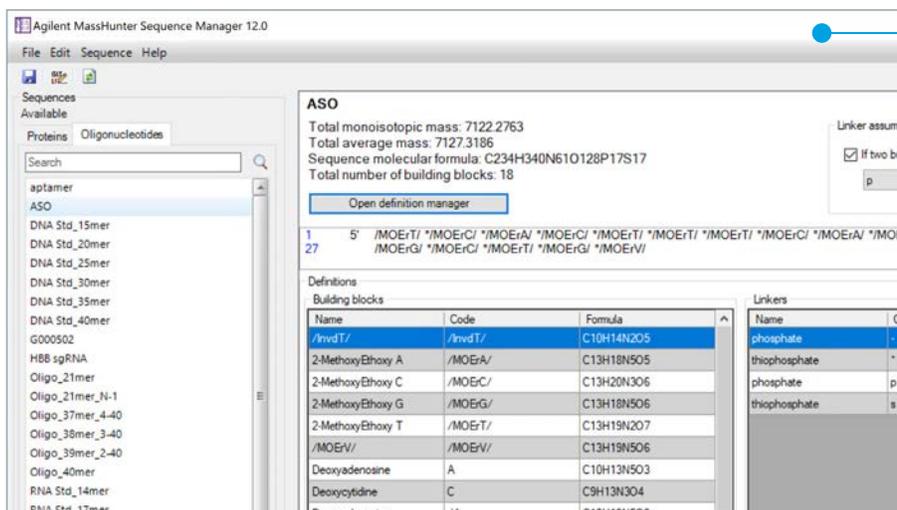
Building Blocks

Code	Name	Formula	Building Block Type
/BaseA/	Adenine	C5H5N5	Base
/BaseC/	Cytosine	C4H5N3O	Base
/BaseG/	Guanine	C5H5N5O	Base
/BaseT/	Thymine	C5H6N2O2	Base
/BaseU/	Uracil	C4H4N2O2	Base
/H/	H		Base
/H2O/	H2O	H2O	Base
/InvdT/	/InvdT/	C10H14N2O5	Nucleoside
/MOEA/	2-MethoxyEthoxy A	C13H18N5O5	Nucleoside
/MOEC/	2-MethoxyEthoxy C	C13H20N3O6	Nucleoside
/MOEG/	2-MethoxyEthoxy G	C13H18N5O6	Nucleoside
/MOET/	2-MethoxyEthoxy T	C13H18N2O7	Nucleoside
/MOEV/	/MOEV/	C13H19N5O6	Nucleoside
/OH/	OH	OH	Base
A	Deoxyadenosine	C10H13N5O3	Nucleoside
C	Deoxycytosine	C9H13N3O4	Nucleoside

New building block code: [] + -

Chemical Data Dictionary

The Chemical Data Dictionary has been enhanced to enable defining oligonucleotide building blocks, linkers, and modifications the way you want them.



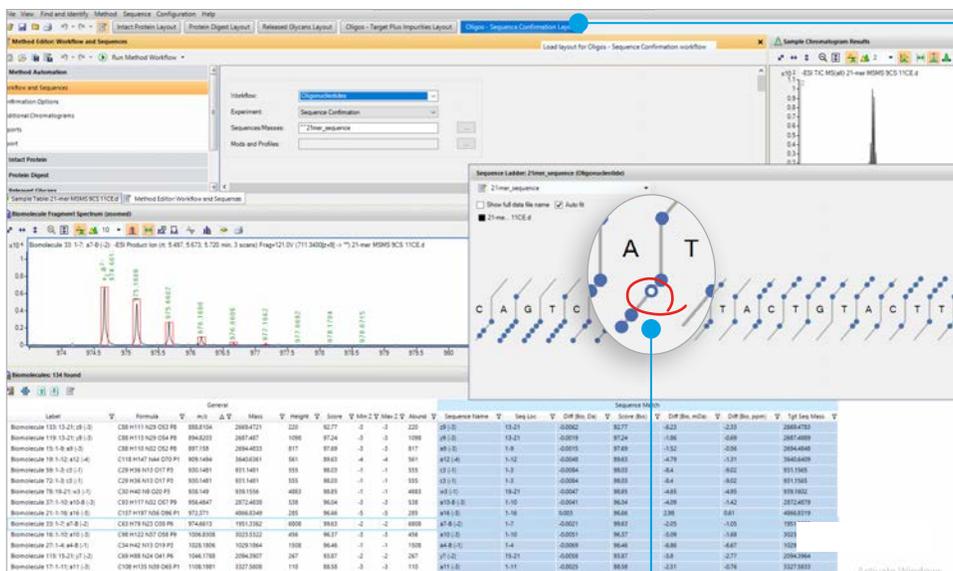
Sequence Manager

The Sequence Manager can take sequence input based on your preferred definitions for building blocks, linkers, and modifications.

Sequence Name	Target/Impurity Name	Target/Impurity Description	Molecular Formula	Mass
Oligo_21mer	CAGTCGATTGACTGTACTTA		C206H261N73O127P20	6408.4
Oligo_21mer	CAGTCGATTGACTGTACTTA(5-Trunc-L)	(5-Trunc-L)	C197H249N70O121P19	6119.4
Oligo_21mer	CAGTCGATTGACTGTACTTA(5-Trunc-L(x2))	(5-Trunc-L(x2))	C187H237N65O116P18	5805.4
Oligo_21mer	CAGTCGATTGACTGTACTTA(5-Trunc-L(x3))	(5-Trunc-L(x3))	C177H225N60O110P17	5476.4
Oligo_21mer	CAGTCGATTGACTGTACTTA(5-Trunc-L(x4))	(5-Trunc-L(x4))	C167H212N58O103P16	5172.4
Oligo_21mer	CAGTCGATTGACTGTACTTA(5-Trunc-L(x5))	(5-Trunc-L(x5))	C158H200N55O97P15	4883.4
Oligo_21mer	CAGTCGATTGACTGTACTTA(5-Trunc-L(x6))	(5-Trunc-L(x6))	C148H188N50O91P14	4554.4
Oligo_21mer	CAGTCGATTGACTGTACTTA(5-Trunc-L(x7))	(5-Trunc-L(x7))	C138H176N45O86P13	4241.4
Oligo_21mer	CAGTCGATTGACTGTACTTA(5-Trunc-L(x8))	(5-Trunc-L(x8))	C128H163N43O79P12	3937.4
Oligo_21mer	CAGTCGATTGACTGTACTTA(5-Trunc-L(x9))	(5-Trunc-L(x9))	C118H150N41O72P11	3633.4
Oligo_21mer	CAGTCGATTGACTGTACTTA(5-Trunc-L(x10))	(5-Trunc-L(x10))	C108H138N36O66P10	3304.4
Oligo_21mer	CAGTCGATTGACTGTACTTA(5-Trunc-L(x11))	(5-Trunc-L(x11))	C98H125N34O59P9	3000.4
Oligo_21mer	CAGTCGATTGACTGTACTTA(5-Trunc-L(x12))	(5-Trunc-L(x12))	C88H113N29O54P8	2687.4
Oligo_21mer	CAGTCGATTGACTGTACTTA(5-Trunc-L(x13))	(5-Trunc-L(x13))	C79H101N26O48P7	2398.4
Oligo_21mer	CAGTCGATTGACTGTACTTA(5-Trunc-L(x14))	(5-Trunc-L(x14))	C69H88N24O41P6	2094.4
Oligo_21mer	CAGTCGATTGACTGTACTTA(5-Trunc-L(x15))	(5-Trunc-L(x15))	C59H76N19O35P5	1765.4

Target Plus Impurities (TPI)

The TPI uses oligonucleotide MS data to identify the full-length product (FLP) as well as any potential impurities.



Perform MS/MS-based sequence confirmation in minutes

Sequence confirmation typically takes up to one week manually. But with BioConfirm software, you can confidently perform MS/MS-based sequence confirmation in minutes with minimal manual steps.

The selected biomolecule for position a7-B is represented on the sequence ladder as an open circle.

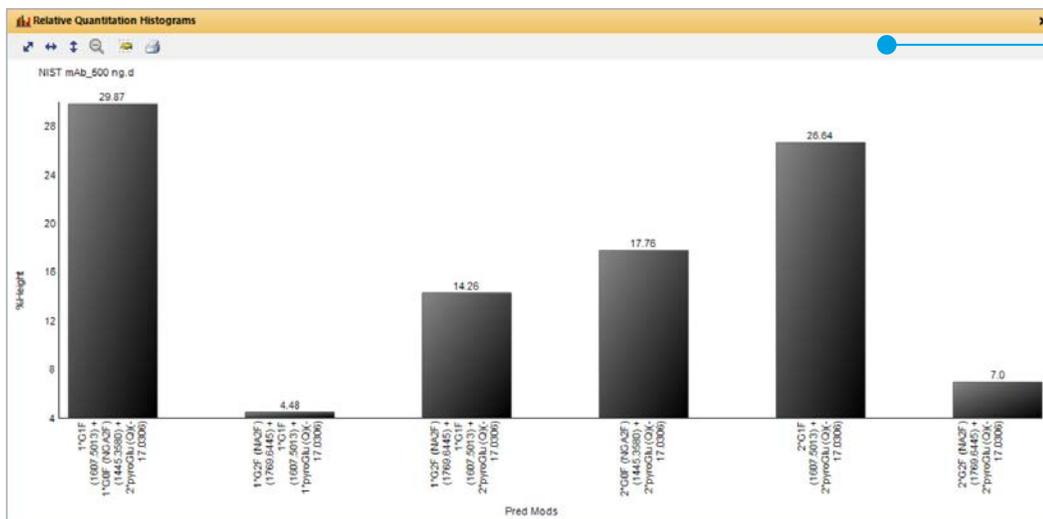
Intact Protein Analysis: Confirm PTMs with Certainty



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

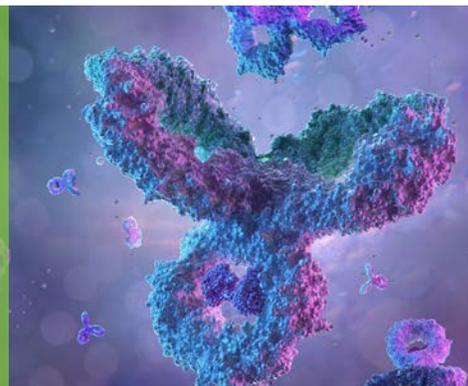
Intact protein analysis in MassHunter BioConfirm also works well with the low-vacuum environment of the Agilent 6545XT AdvanceBio LC/Q-TOF mass spectrometer. It allows more proteins to get through for greater sensitivity.

Predicted modifications

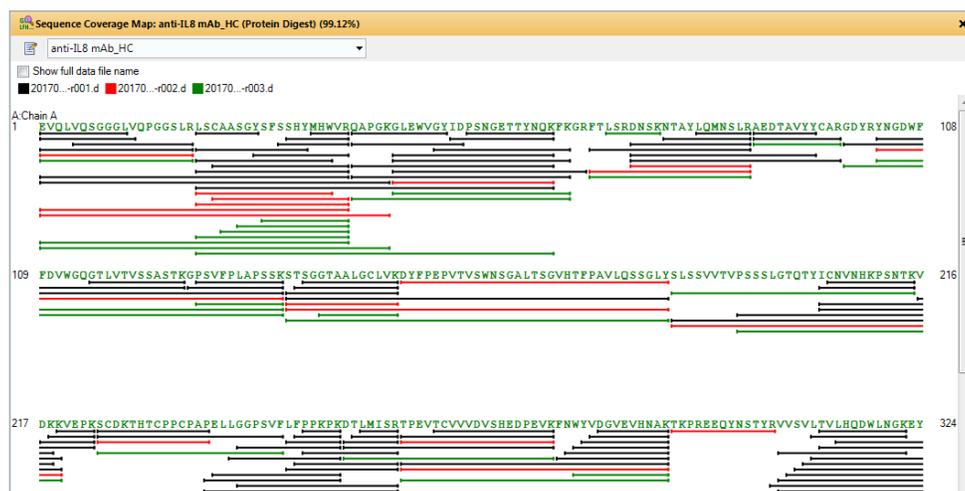


Relative quantitative results of glycoforms are displayed as histogram plots, allowing you to monitor a process.

Peptide Mapping: Be Sure of Your Sequences



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. 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.



Sequence Coverage Map

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

Location	Pred Mods	File	%Quant (Height)	Height
M255 [B/D]	Oxidation (M)	NIST mAb_peptide m	14.24	121589
Sequence	Pred Mods	Use for %Quant	Height	
LMISR		<input type="checkbox"/>	617051	
DTLMISR		<input checked="" type="checkbox"/>	723330	
TLMISR		<input type="checkbox"/>	376143	
TLMISR		<input type="checkbox"/>	278204	
LMISR		<input type="checkbox"/>	220787	
DTLMISR	Oxidation (M) 4	<input checked="" type="checkbox"/>	121589	
DTLMISR		<input checked="" type="checkbox"/>	8898	
TLMISR	Oxidation (M) 3	<input type="checkbox"/>	5981	

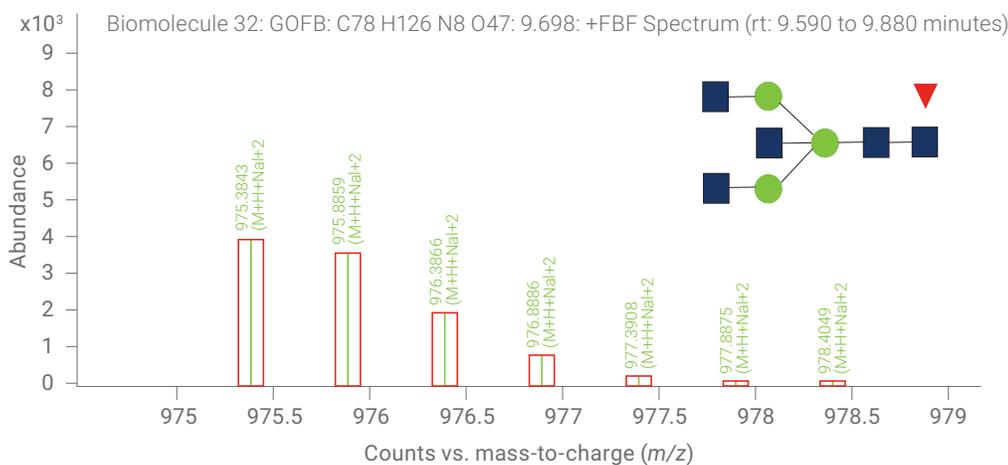
Relative quant results

Perform relative quantitation of modified and unmodified peptides.

Released Glycan Profiling: Improve Sensitivity



Glycans are a challenging PTM to characterize given the diversity in their composition and the need for chemical tagging to ensure analytical sensitivity. 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 software tools. Learn more about N-glycan analysis at www.agilent.com/chem/glycan-sampleprep



The released glycans results include spectra annotated with the glycan structure and theoretical isotope distribution. The Agilent Find-by-Formula algorithm takes advantage of the glycan tendency to ionize with multiple adducts, resulting in finding species, such as M+H+Na, and giving greater specificity.

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.

Sample Information

(C)SampleName	(C)SampleName	(C)DateAcq	(C)DateAcq
(C)SampleID	(C)SampleID	(C)AcqTime_Limit	(C)AcqTime_Limit
(C)InstrumentName	(C)InstrumentName	(C)Acquisition_Acquisition	(C)Acquisition_Acquisition
(C)MSType	(C)MSType	(C)Version_Active	(C)Version_Active
(C)AcquisitionName	(C)AcquisitionName	(C)Method	(C)Method
(C)SamplePath	(C)SamplePath	(C)Acquisition_Acquisition	(C)Acquisition_Acquisition
(C)SampleName	(C)SampleName	(C)SampleName	(C)SampleName
(C)Acquisition	(C)Acquisition	(C)Acquisition	(C)Acquisition
(C)Acquisition	(C)Acquisition	(C)Acquisition	(C)Acquisition

Deconvoluted Spectra

Two mass spectra plots are shown side-by-side. The left plot is labeled 'Full Range' and the right plot is labeled 'Zoomed'. Both plots show relative intensity versus m/z, with several peaks labeled with their m/z values.

Biomolecule Summary

Sample	Seq Name	Mass	Top Mass	Diff (Da)	Area (MS)	Height (MS)	Retention	Height	MS/MS	Final Mass
1	NS006	14582	17682	3	43586	17482	1.57	1.57	100	14582

Settings Panel

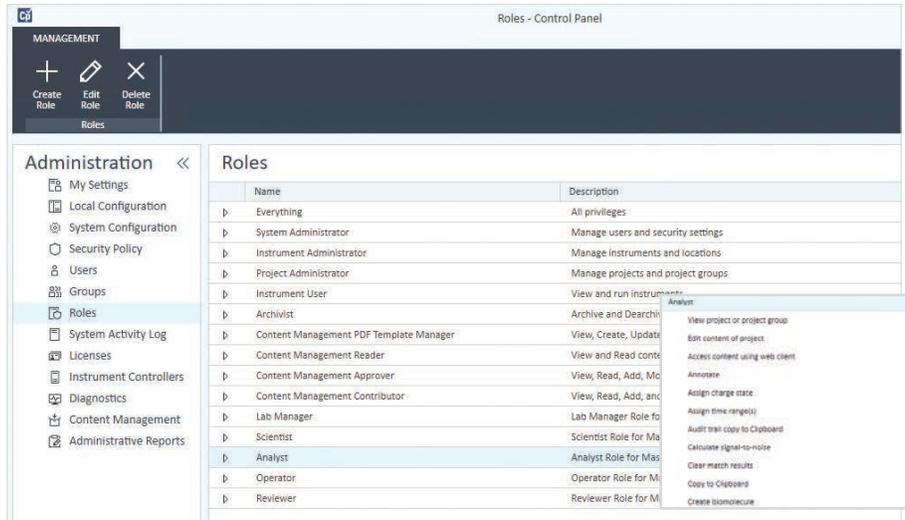
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- Font: [Dropdown]
- Color: Black
- Field Format: [Dropdown]
- Format: [Dropdown]
- Control: [Dropdown]
- ID: Textbox1
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Annotations:

- Blue dot: Add your company logo
- Blue dot: Place tables, chromatograms, and spectra exactly where you want them
- Blue dot: Create PDF reports quickly

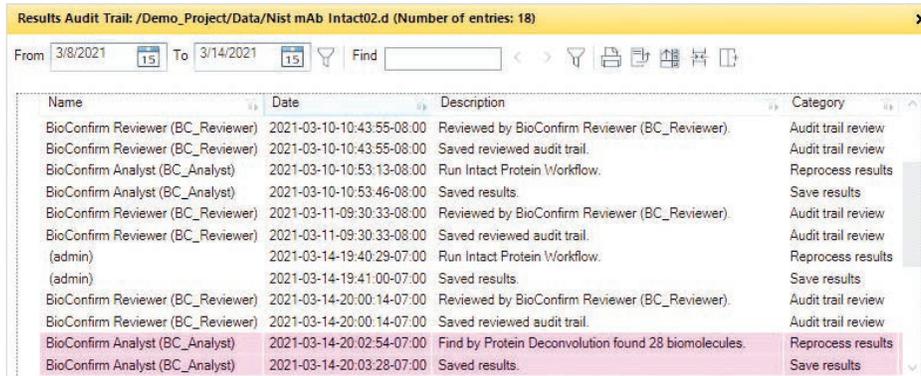
Compliance-ready features keep you in control

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



Control Panel

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



Audit trails

Audit trails make reviewing easy by highlighting entries that require review. Tamper detection is automatically included using checksums.



Which version of BioConfirm software should I choose?

MassHunter BioConfirm software 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 find the version of BioConfirm biopharmaceutical software for LC/Q-TOF that is best for your lab.

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

○ = Optional feature

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.

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