

Agilent AssayMAP Bravo Platform

Automated protein and peptide sample preparation for mass spec analysis



Superior reproducibility with automated sample prep

The Agilent AssayMAP platform is a powerful yet easy-to-use automated platform for protein and peptide sample preparation. The system consists of a precision liquid handler with 96 syringes, packed-bed cartridges, and intuitive software with pre-optimized sample prep protocols. Together, these components enable sample prep to be performed in a parallel on 1 to 96 samples with all the advantages of chromatography.

- · Increase workflow reproducibility by reducing sample prep variability
- Reduce tedious manual steps and free up time for more value-added work
- · Increase throughput without increasing variability or labor cost
- · Seamless transfer from person-to-person, lab-to-lab and site-to-site
- Obtain higher yields from your samples in a lower volume



Three key elements help you succeed

- Two sizes of chromatography cartridges with a wide range of chemistries
- Precisely controlled liquid flow for highly reproducible binding and elution
- Intuitive software combined with laboratory-tested protocols designed for simplicity

A single platform for multiple biopharma and proteomics workflows

The AssayMAP platform is an out of the box sample prep solution for protein/peptide characterization and quantification workflows such as intact mass analysis, peptide quantification, peptide mapping, post translational modifications, and many more.

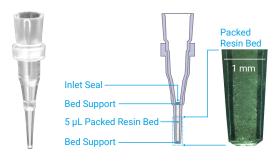
The system includes pre-optimized protocols for affinity purification, enzymatic digestions, reversed-phase peptide/protein cleanup, phosphopeptide enrichment, peptide fractionation, and standard liquid handling.

Diverse cartridge chemistries

AssayMAP cartridges are available in 5 and 25 μ L packed bed formats. The cartridge options include:

- Protein A resin wide pore (PA-W)
- Protein G resin wide pore (PG-W)
- Streptavidin resin wide pore (SA-W)
- Reversed-phased resin wide pore (RP-W)
- Reversed-phase resin small pore (PR-S)
- Reversed-phase resin small pore (C18)
- Strong cation exchange resin small pore (SCX)
- Titanium dioxide resin small pore (TiO2)
- Fe(III)-NTA resin wide pore (Fe(III)-NTA)

Please use Cartridge Selection Guide, 5991-4863EN for additional information such as cartridge binding capacities.



Precise flow control

Precisely controlled liquid flow across the cartridge resin bed is guaranteed by near zero dead volume, positive displacement probe syringes, resulting in unparalleled binding and elution performance in a single pass.

Ninety-six probe syringes are housed on the AssayMAP multichannel head, allowing you to run 1 to 96 samples in a single run. The probes can also mount pipette tips for standard liquid handling operation.

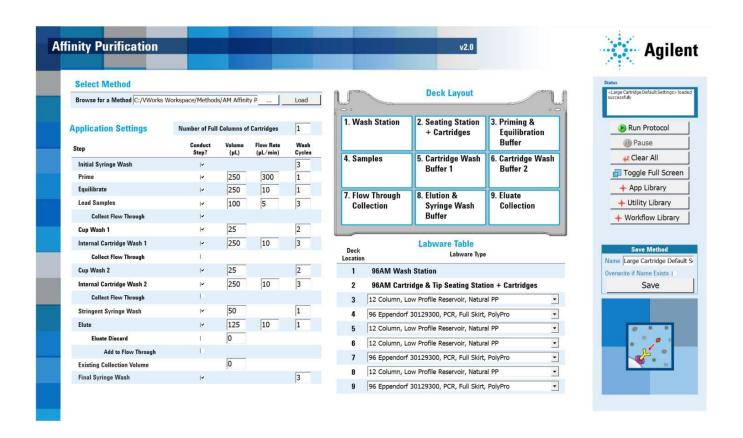


Software to address a wide range of workflows

Protein Sample Prep Workbench: APP LIBRARY **Affinity Purification v2.0** This is the recommended Affinity Purification application. Enrich for target molecules using Protein A, Protein G, or user-defined affinity cartridges. All reagents flow from the cup to the tip of the cartridges in dispense mode. Using AssayMAP Quick Start Guide Bravo and Cartridges. Fractionation v1.1 Bind, wash and elute in up to six fractions with different stepwise elution buffers. Using AssayMAP Bravo and Cartridges. Quick Start Guide Calculator IMAC Cartridge Customization v1.1 12. Strip existing metal from cartridges and charge with metal of choice. Using AssayMAP Bravo and Fe(III)-NTA cartridges. Quick Start Guide 70 Calculator Immobilization v2.0 Immobilize an affinity ligand to create a user-defined affinity cartridge. Using AssayMAP Bravo and Cartridges. Quick Start Guide Calculator In-Solution Digestion: Single-Plate v1.2 Ouick Start Guide **On-Cartridge Reaction v1.1** Conduct a reaction using samples immobilized on affinity cartridges, and a temperature controlled reagent aspirated through the cartridges. Using AssayMAP Bravo and Cartridges Ouick Start Guide Peptide Cleanup v3.0 This is the recommended Peptide Cleanup application. Clean peptides from complex digests. All reagents flow from the cup to the tip of the cartridges in dispense mode. Mixing steps are included. Using AssayMAP Bravo and Quick Start Guide Calculator Phosphopeptide Enrichment v2.1 Enrich for phosphopeptides from complex samples. Using AssayMAP Bravo and Fe(III) NTA or TiO, cartridges. Ouick Start Guide Protein Cleanup v2.0 Remove unwanted salts and buffers from protein samples by reversed-phase separation. Using AssayMAP Bravo and RP-W Cartridges. Ouick Start Guide Calculator

Pre-optimized protocols to help you succeed quickly

The Protein Sample Prep Workbench is a software interface guiding you through laboratory-tested protocols. Easily create, adjust, and save your methods by using the intuitive navigations split into three sections. The applications settings list the protocol sequence and parameters, the deck layout provides instructions on where to load samples, reagents, and consumables on deck, and the labware table defines what labware to use for different deck positions. With this interface, standard protocols become easy to set up and run for anyone without prior automation experience.



Automated protein sample preparation

Whether you are performing routine protein mass confirmation, or faced with the challenge of hard-to-detect peptides, your workflow begins with the automated AssayMAP Protein and Peptide Sample Preparation platform. With increased consistency, reliability, efficiency, and walk-away time; the AssayMAP system helps you to obtain more consistent results with less effort.



Comprehensive solutions for protein characterization

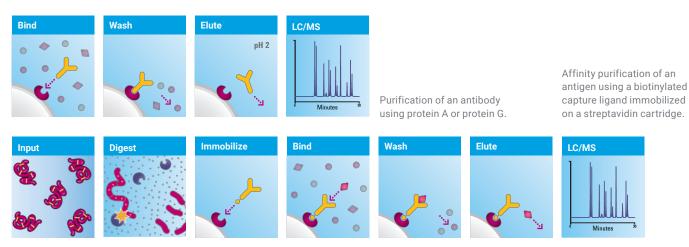
The AssayMAP system is also part of the Agilent Solutions for protein quantitation and characterization which includes Agilent LCs and columns for separation, protein and peptide detection with Agilent mass specs, and protein and peptide sequence confirmation with MassHunter BioConfirm.



Streamlined sample preparation for a growing range of applications

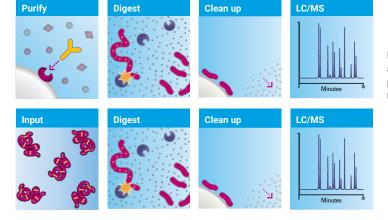
Protein and peptide quantitation

Accurate and reproducible quantiation is a critical part of mutliple steps in the development of biotherapeutics and identification of biomarkers. The AssayMAP Bravo automates the purification of target proteins and peptides from either native or digested samples such as cell culture supernatants, cell lysates, and serum. Purification is quantitative, so you can easily determine the concentration of the target protein in the original sample and minimize the sample consumption. AssayMAP technology enables to move from manual, error-prone and laborious procedures to an automated, highly reproducible, and scalable workflow. Currently available affinity cartridges include streptavidin, protein A, and protein G.



Protein and peptide characterization

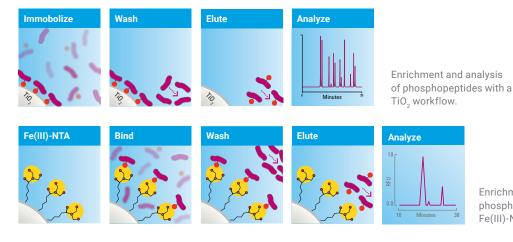
Detailed characterization is another critical aspect of biotherapeutic development and proteomic studies. These workflows often require a complex series of steps that typically include digestion with enzymes such as trypsin, cleanup of the resulting peptide mixture, and sometimes fractionation before analysis on an LC/MS system. Depending on the workflow, there is sometimes an affinity purification of the analytical target before the series of steps. Examples of the workflows that use this type of workflow in biotherapeutic and proteomic research include peptide mapping and post translational modifications. We have developed AssayMAP protocols that provide end-to-end solutions for these workflows.



Purification, proteolysis, and desalting of a target protein using the peptide mapping workflow.

Phosphopeptide enrichment

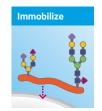
Phosphorylation is a common post translational modification that is used in biomarker discovery and mechanism of action studies. Due to the low abundant nature of phosphorylation, you need a strategy to capture and enrich the phosphopepetides before you analyze them with LC/MS. The AssayMAP workflow gives you multiple automated solutions that reproducibly enrich phosphopeptides using either the Fe(III)-NTA, TiO2, or immobilized anti-phophopeptide antibodies. The AssayMAP platform allows you to use any or all of these techniques to analyze your sample.



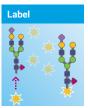
Enrichment and analysis of phosphopeptides with an Fe(III)-NTA workflow.

N-glycan sample preparation

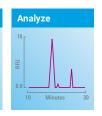
The AssayMAP protocol combined with optimized reagents from ProZyme kits reduce N-Glycan sample prep from three days to four hours. The target glycoprotein is denatured, immobilized on a cartridge, then digested with PNGase F, which specifically releases N-glycans from the target protein. Next, the released glycans are chemically labeled with a number of dye choices including a dual function dye that enables both mass spectrometry and fluorescence detection. A cleanup cartridge is used before analysis to minimize interference from unincorporated dye.











Glycoprotein immobilization, glycan release, glycan labeling, and cleanup.

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