

ProteoMiner™ Protein Enrichment Technology

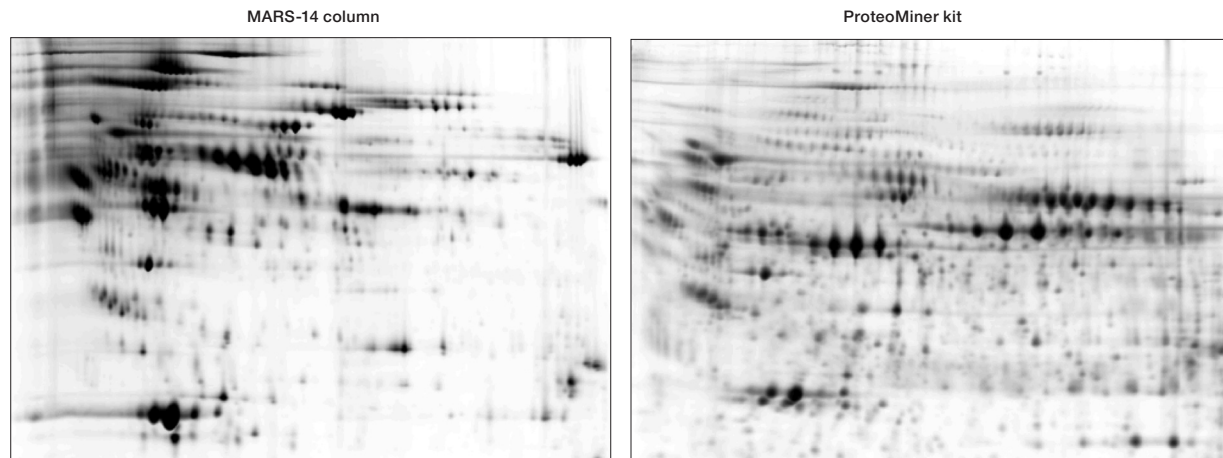
Digging Deeper in  
the Proteome



## Detect More Proteins With ProteoMiner Technology Than With Immunodepletion

### ProteoMiner Technology vs. an Agilent MARS-14 Column

A side-by-side comparison of the ProteoMiner protein enrichment kit and the Agilent MARS-14 column was performed using a plasma sample. Following treatment with either the ProteoMiner kit or the MARS column, the samples were analyzed by two-dimensional (2-D) gel electrophoresis. The sample treated with the ProteoMiner kit showed many more protein spots than did the sample treated with the MARS-14 column. These results suggest that the ProteoMiner kit is a superior product for sample preparation prior to low-abundance protein studies.

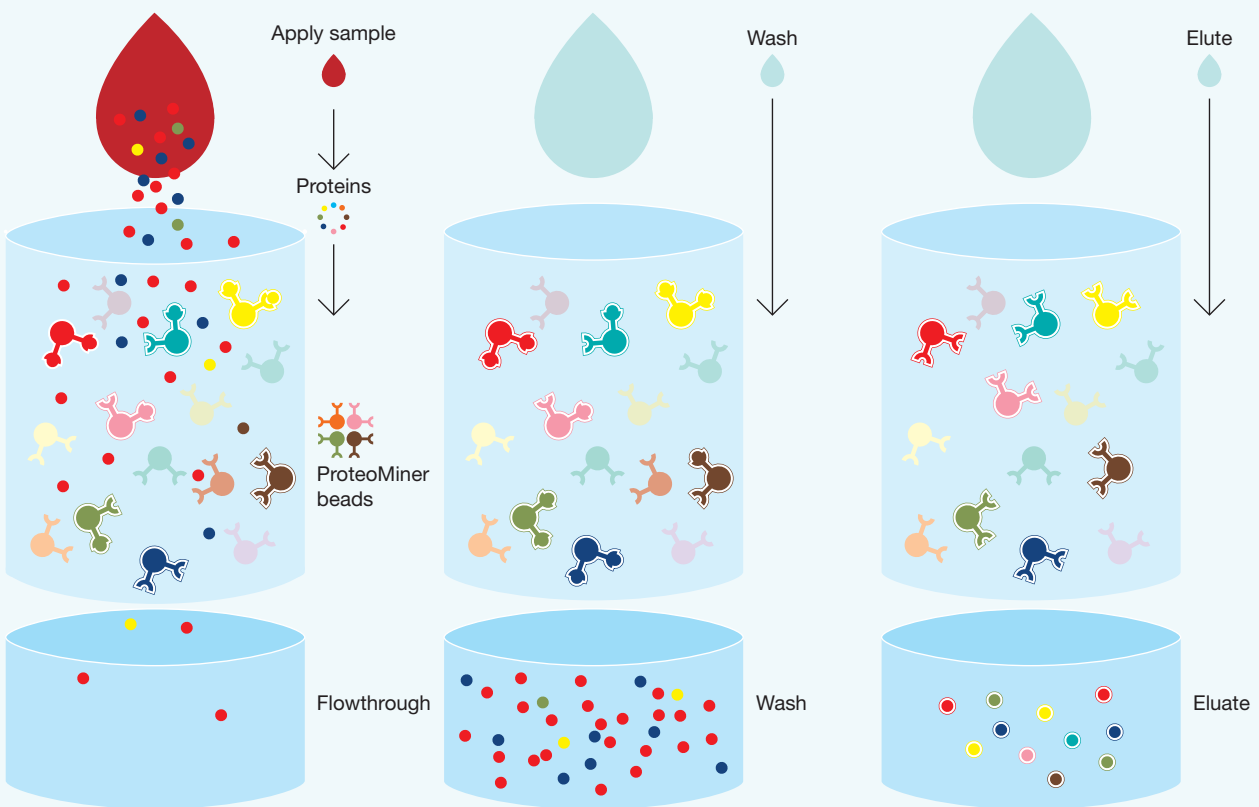


**Results of comparison of two protein sample preparation methods.** 2-D gel electrophoresis was performed on a plasma sample depleted with an Agilent MARS-14 column (left) and with the ProteoMiner kit (right). A substantially greater number of proteins were detected after treatment with the ProteoMiner kit. Data provided by AstraZeneca.

# ProteoMiner Protein Enrichment Kits

## Uncover Low-Abundance Proteins

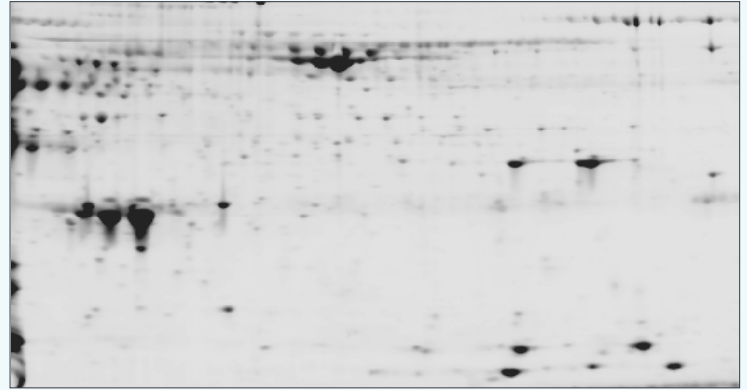
ProteoMiner protein enrichment technology is a novel sample preparation tool used to compress the dynamic range of protein concentrations in complex biological samples. The presence of high-abundance proteins in complex biological samples (for example, albumin and IgG in serum or plasma) makes the detection of medium- and low-abundance proteins extremely challenging. ProteoMiner protein enrichment kits provide a method for overcoming this challenge, allowing the exploration of the entire proteome.



ProteoMiner technology is based on the interaction of complex protein samples with a large, highly diverse library of hexapeptides bound to chromatographic supports. In theory, each unique hexapeptide binds to a unique protein sequence. Because the bead capacity limits binding capacity, high-abundance proteins quickly saturate their ligands (red and yellow beads) and excess protein is washed out during the procedure. In contrast, low-abundance proteins are concentrated on their specific ligands (pink and teal beads), thereby decreasing the dynamic range of proteins in the sample. When analyzed in downstream applications, the number of proteins detected is dramatically increased.



Untreated



Treated

**The ProteoMiner protein enrichment kit improves resolution and spot counts in 2-D gels.** In an untreated sample, albumin and other high-abundance proteins dominate the gel, obscuring signals from less abundant proteins; on a gel generated using an equal amount of total protein from a treated serum sample, however, resolution is dramatically improved, and a greater number of protein spots are visualized.

### Key Benefits of ProteoMiner Technology

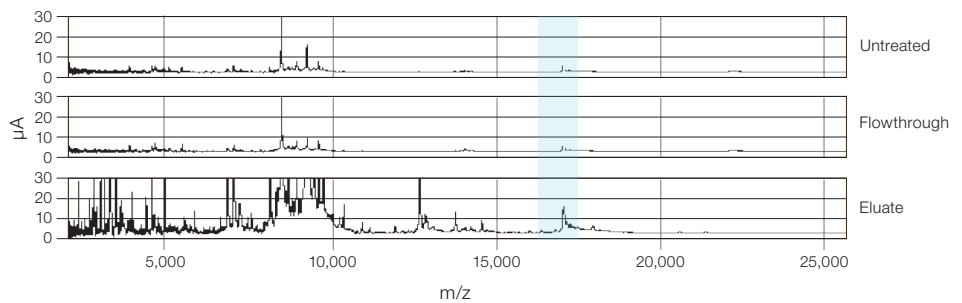
- Decreases the amount of high-abundance proteins without immunodepletion; prevents co-depletion of proteins bound to high-abundance proteins
- Enriches and concentrates low-abundance proteins that cannot be detected through traditional methods
- Compresses the dynamic range of protein concentration in a variety of samples and is not dependent on a predefined set of antibodies as are immunodepletion products
- Can be used for differential expression analysis
- Is compatible with all major downstream protein analysis techniques

### Convenient Format

- Spin column format is easy to use
- Onetime use avoids potential for contamination from carryover between samples
- All necessary columns and reagents provided in kits

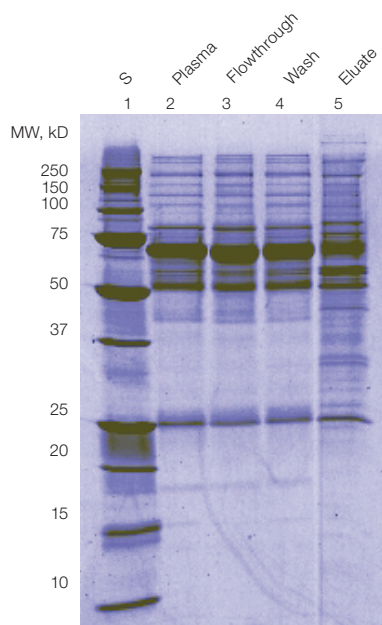
### Discover More Proteins

Whether you use 1-D or 2-D gel electrophoresis, chromatography, surface-enhanced laser desorption/ionization (SELDI), or other mass spectrometry technologies for your downstream analysis, ProteoMiner kits provide a tool for digging deeper in the proteome.



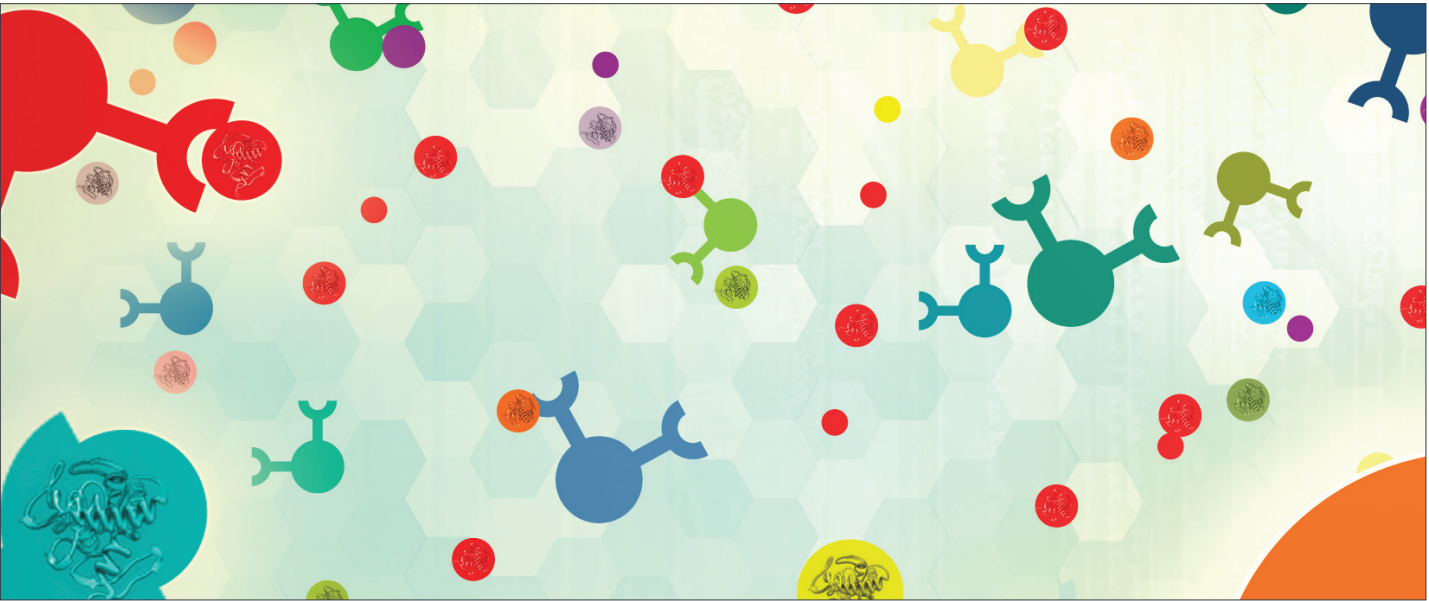
### The ProteoMiner protein enrichment kit improves peak counts in SELDI analysis.

ProteinChip® SELDI system analysis with CM10 array of untreated plasma reveals 170 peaks, while plasma treated with the ProteoMiner protein enrichment kit (eluate) yields 263 peaks, or 55% more. One peak (shaded) was selected to show the enrichment experienced after the sample was treated with the ProteoMiner kit.



### SDS-PAGE analysis of plasma treated with the ProteoMiner protein enrichment kit.

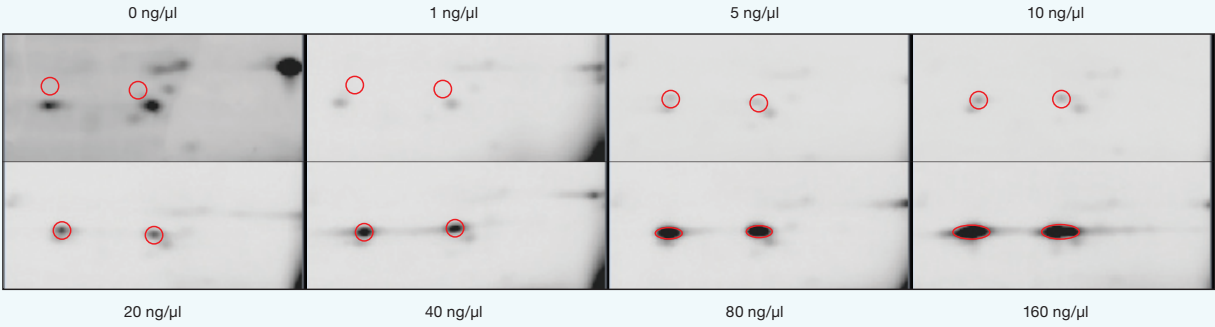
A plasma sample (1 ml) was treated with the ProteoMiner protein enrichment kit, and 75 µg protein each from the original sample (lane 2), flowthrough (lane 3), wash (lane 4), and elution fractions was analyzed by SDS-PAGE. S, Precision Plus Protein™ standard.



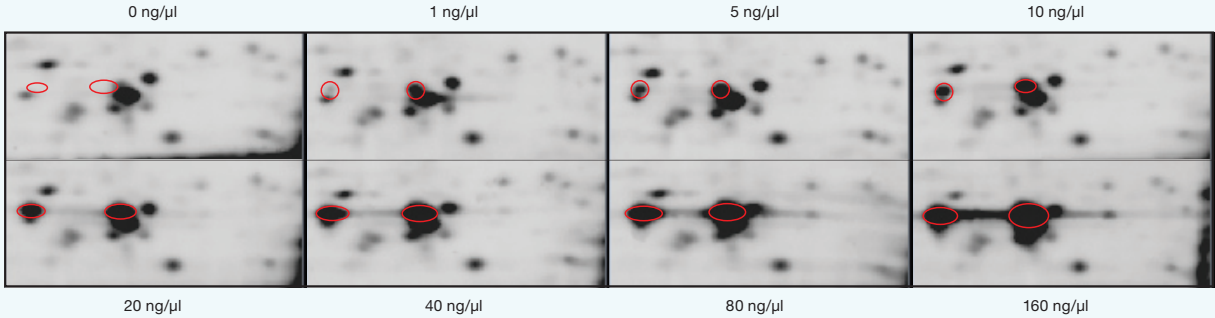
**Enhance Potential for Biomarker Discovery With ProteoMiner Technology**

In biomarker discovery studies, accurate quantitation of low-abundance proteins in both disease and control samples is critical. ProteoMiner technology preserves quantitative information and enriches low-abundance proteins to make biomarker discovery easier and more likely.

**Untreated**



**Treated**



**ProteoMiner technology enriches proteins while maintaining the relative expression levels of low- to medium-abundance proteins.** A series of plasma samples were spiked with different concentrations of serum amyloid A (SAA), left untreated (top) or treated with the ProteoMiner protein enrichment kit (bottom), and analyzed by 2-D gel electrophoresis. Relative intensities of the SAA spots (circled) are proportional to the amount of SAA added both before and after treatment with the ProteoMiner kit. The SAA spots are detected at a lower concentration (~5 ng/μl) following treatment with the ProteoMiner kit vs. that seen in the untreated sample (~10 ng/μl), indicating that the ProteoMiner kit successfully enriched SAA.

### Multiple Kits Available

To provide flexibility for use with multiple downstream applications, two kit formats are available — the ProteoMiner protein enrichment kit and the ProteoMiner sequential elution kit. These kits have been optimized for use with serum and plasma samples, but they may be used with other sample types with at least 50 mg total protein available.

#### ProteoMiner Protein Enrichment Kit

The ProteoMiner protein enrichment kit provides columns and all necessary reagents for accessing low-abundance proteins in a variety of biological samples and is compatible with the majority of downstream proteomics applications. The full-size kit can be used to process up to ten samples, while the ProteoMiner introductory kit can be used to process two samples.

- Utilizes single elution reagent in simple, easy-to-perform process
- Provides maximum compatibility with downstream applications

#### ProteoMiner Sequential Elution Kit

The ProteoMiner sequential elution kit is available for researchers who wish to elute their proteins into multiple fractions to detect additional proteins. This kit has been optimized for SELDI experiments and provides columns and reagents for processing up to ten samples (reagents are not compatible for direct use with 2-D gel electrophoresis).

- Multiple elution reagents sequentially elute proteins based on different properties
- Fractionates proteins to improve detection and resolution of proteins for SELDI analysis

For more information, request bulletin 5632, or visit us on the Web at [www.bio-rad.com/proteominer/](http://www.bio-rad.com/proteominer/)

Low-abundance protein enrichment services are available through Bio-Rad's BioMarker Research Centers. To learn more, contact your local Bio-Rad representative or send an e-mail to [BRC@bio-rad.com](mailto:BRC@bio-rad.com).

### Bibliography

Boschetti E et al., Reduction of the concentration difference of proteins in biological liquids using a library of combinatorial ligands, *Electrophoresis* 26, 3561–3571 (2005)

Boschetti E et al., Reducing protein concentration range of biological samples using solid-phase ligand libraries, *J Chromatogr B Biomed Sci Appl* 833, 33–40 (2006)

Sennels L et al., Proteomic analysis of human blood serum using peptide library beads, *J Proteome Res* 4055–4062 (2007)

### Ordering Information

Catalog #	Description
163-3000	<b>ProteoMiner Protein Enrichment Kit</b> , 10 preps, includes 10 spin columns, wash buffer, elution reagents, collection tubes
163-3001	<b>ProteoMiner Introductory Kit</b> , 2 preps, includes 2 spin columns, wash buffer, elution reagents, collection tubes
163-3002	<b>ProteoMiner Sequential Elution Kit</b> , 10 preps, includes 10 spin columns, wash buffer, 4 sequential elution reagents, collection tubes
163-3003	<b>ProteoMiner Sequential Elution Reagents</b> , 10 preps, includes reagents only (columns not included), to be used with catalog #163-3000



Agilent is a trademark of Agilent Technologies, Inc.

The SELDI process is covered by US patents 5,719,060, 6,225,047, 6,579,719, and 6,818,411 and other issued patents and pending applications in the US and other jurisdictions.

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