High-Select™ Top14 Abundant Protein Depletion Resin

Catalog Numbers A36369, A36370, A36371, and A36372

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Contents

Cat. No.	Product	Contents	Storage
A36369	High-Select™ Top14 Abundant Protein Depletion Mini Spin Columns	6 columns Each column contains 400 µL of a 50% slurry in 10mM PBS, 0.02% sodium azide, pH 7.4	Store at 4°C. Do not freeze.
A36370	High-Select™ Top14 Abundant Protein Depletion Mini Spin Columns	24 columns Each column contains 400 µL of a 50% slurry in 10mM PBS, 0.02% sodium azide, pH 7.4	
A36371	High-Select™ Top14 Abundant Protein Depletion Midi Spin Columns	10 columns Each column contains 1,000 µL of a 50% slurry in 10mM PBS, 0.02% sodium azide, pH 7.4	
A36372	High-Select™ Top14 Abundant Protein Depletion Resin	50 mL 50% slurry in 10mM PBS, 0.02% sodium azide, pH 7.4	

Product description

Thermo Scientific™ High-Select™ Top14 Abundant Protein Depletion Resin is used for single-step removal of 14 high-abundant proteins from 10 µL (mini format) or 100 µL (midi format) of human serum or plasma. Alternatively, researchers may customize with the bulk resin to suit experimental needs. The resin uses highly specific immobilized antibodies for protein removal, providing minimal nonspecific interactions with other proteins.

Plasma proteome studies are limited by the vast dynamic range (10 orders of magnitude) of protein abundance. Many proteins of interest are present at low amounts in plasma and are difficult to detect in the presence of high-abundant proteins. The proteins listed in the table below constitute ~95% of all plasma proteins. The High-Select™ Top14 Abundant Protein Depletion Resin removes >95% of these 14 high abundant proteins from serum or plasma samples and enables the identification and quantitation of low-abundant proteins in samples by mass spectrometry (MS).

Table 1 Top 14 proteins removed by High-Select™ Top14 Abundant Protein Depletion Resin.

- Albumin
- IgA
- IgD
- IgE
- IgG
- IgG (light chains)
- IgM
- Alpha-1-acid glycoprotein
- Alpha-1-antitrypsin
- Alpha-2-macroglobulin
- Apolipoprotein A1
- Fibrinogen
- Haptoglobin
- Transferrin

Additional information

- The depletion spin columns contain a storage solution used as a dilution and binding buffer for direct processing of 10 µL for the mini format or 100 µL for the midi format of serum or plasma.

  Note: No other additions or solvent exchanges are required before protein depletion.
- The mini depletion spin columns can process a maximum of 10 µL of human serum or plasma. The midi depletion spin columns can process a maximum of 100 µL of human serum or plasma. For serum samples containing abnormally high amounts of albumin or IgG, the sample load may need to be reduced.
- The depletion spin columns are designed for single use. **Do not reuse the resin.**
- The depletion resin is designed for use with human serum or plasma samples and has not been tested on any other species. Alternative human biological fluids (e.g., cerebrospinal fluid or amniotic fluid) may be used with this product but may require optimization.
- The depletion resin allows the user to customize the fill of resin depending on experimental needs. This resin is designed to only be used once with sample. **Do not reuse the resin.**

Additional materials required

- Microcentrifuge and centrifuge capable of operating at 1,000 × *g*
- 2 mL collection tubes
- 15 mL collection tubes
- End-over-end mixer

Remove high-abundance top 14 proteins

The concentration of serum/plasma proteins can vary widely depending on the origin of the sample. Each mini column (200 µL of antibody resin bed) is optimized to bind up to 10 µL (600 µg) of serum or plasma and each midi column (500 µL of antibody resin bed) binds up to 100 µL (6,000 µg) of serum or plasma. For best results, optimize the ratio of sample to slurry volume for each specific application.

Remove high-abundance top 14 proteins in mini format

1. Equilibrate the depletion spin column to room temperature.
2. Remove the column screw cap and add up to 10 µL of sample directly to the resin slurry in the column.
3. Cap the column and invert the column several times until the resin is completely homogenous in solution.
4. Incubate the mixture in the column with gentle end-over-end mixing for 10 minutes at room temperature. Make sure the sample mixes with the resin during incubation period. Alternatively, gently vortex every few minutes.
5. After incubation, snap off the bottom closure and loosen the top cap. Place the mini column into a 2 mL collection tube and centrifuge at 1,000 × *g* for 2 minutes.
6. Discard the column containing the resin.
7. Filtrate contains sample with albumin, IgG, and other abundant proteins removed. Use for further processing or store at -20°C for later use. The depleted sample will be in 10 mM PBS and 0.02% sodium azide, pH 7.4.

   Note: Sample processing will depend on the type of downstream analysis and may require buffer exchange, lipids and other metabolite removal and/or concentration for 2D gel electrophoresis and MS analysis. Use Thermo Scientific™ Pierce™ Protein Concentrators for buffer exchanging and/or concentrating.

Remove high-abundance top 14 proteins in midi format

1. Equilibrate the depletion spin column to room temperature.
2. Remove the column screw cap and add up to 100 µL of sample directly to the resin slurry in the column.
3. Cap the column and invert the column several times until the resin is completely homogenous in solution.
4. Incubate the mixture in the column with gentle end-over-end mixing for 10 minutes at room temperature. Make sure the sample mixes with the resin during incubation period. Alternatively, gently vortex every few minutes.
5. After incubation, snap off the bottom closure and loosen the top cap. Place the mini column into a 15 mL collection tube and centrifuge at 1,000 × *g* for 2 minutes.
6. Discard the column containing the resin.
7. Filtrate contains sample with albumin, IgG, and other abundant proteins removed. Use for further processing or store at -20°C for later use. The depleted sample will be in 10 mM PBS and 0.02% sodium azide, pH 7.4.

   Note: Sample processing will depend on the type of downstream analysis and may require buffer exchange, lipids and other metabolite removal and/or concentration for 2D gel electrophoresis and MS analysis. Use Thermo Scientific™ Pierce™ Protein Concentrators for buffer exchanging and/or concentrating.

Troubleshooting

Observation	Possible cause	Recommended action
Albumin or IgG were not completely removed.	Sample exceeded binding capacity.	Reduce amount of sample processed.
	Incomplete binding.	Increase incubation time.
	Sample was not mixed during incubation.	Mix the sample with resin with gentle end-over-end mixing and make sure that the sample is mixing with the resin during the incubation period.

Limited product warranty

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