Multiple Affinity Removal Spin Cartridge Human-7, 0.45 ml

Part No. 5188-6408
For depletion of seven high-abundance proteins from human plasma samples

Instructions
Version A1, December 2018
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Notices

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In this Guide...

This document describes how to use the Multiple Affinity Removal Spin Cartridge Human-7, 0.45 ml to chromatographically remove seven interfering high-abundance proteins from human plasma samples prior to LC/MS or electrophoretic analysis of the samples.

1 Before You Begin

This chapter contains information (such as required reagents and equipment) that you should read and understand before you start an experiment.

2 Instructions

This chapter describes the protocol for chromatographic removal of the targeted proteins from human plasma samples and includes troubleshooting information.

3 Reference

This chapter contains reference information including cartridge specifications and a list of related products.
What’s New in Version A1

• Updates to description of needles included with reagent kits (see Table 1 on page 7, Table 6 on page 21)

• Correction to part numbers for spin cartridge Starter Reagent Kit, p/n 5188-5254, syringes, p/n 5188-5250, and mRP-C18 desalting column, p/n 5188-5231 (see Table 1 on page 7, Table 6 on page 21)

• Updates to Technical Support contact information (see page 2)
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Before You Begin

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Make sure you read and understand the information in this chapter and have the necessary equipment and reagents listed before you start an experiment.
Safety Considerations

When preparing biological samples using Agilent Multiple Affinity Removal Spin Cartridges, follow general guidelines for handling biological materials and wear protective eyewear and gloves.

Materials Required

Ordering information for the Multiple Affinity Removal Spin Cartridge and additional materials used in the protocol is provided in Table 1 (materials from Agilent) and Table 2 (materials from external suppliers.)

NOTE

For higher capacity Multiple Affinity Removal Devices for human plasma, and if automated immunodepletion is needed, refer to the Multiple Affinity Removal Columns (part numbers 5188-6409 and 5188-6410) for use with HPLC instrumentation. See www.agilent.com for more information.

<table>
<thead>
<tr>
<th>Agilent Part number</th>
<th>Product name</th>
<th>Description</th>
</tr>
</thead>
<tbody>
<tr>
<td>5188-6408</td>
<td>Multi Affinity Removal Spin Cartridge Human-7, 0.45 mL, 1 each</td>
<td>Removes albumin, IgG, IgA, transferrin, haptoglobin, antitrypsin, and fibrinogen from human plasma samples. Includes spin cartridge, 2 Luer-Lok adapters, 1 screw cap, and 1 plug</td>
</tr>
<tr>
<td>5185-5987</td>
<td>Buffer A, 1 L</td>
<td>Ready-to-use, optimized buffer for loading, washing, and equilibrating spin cartridge</td>
</tr>
<tr>
<td>5185-5988</td>
<td>Buffer B, 1 L</td>
<td>Ready-to-use, optimized buffer for elution of bound proteins from spin cartridge</td>
</tr>
<tr>
<td>5185-5990</td>
<td>Spin filters, 0.22 µm, 1 pack of 25</td>
<td>For sample cleanup before loading spin cartridge</td>
</tr>
<tr>
<td>5185-5991</td>
<td>Concentrators, 5 kDa MWCO, 1 pack of 25</td>
<td>For concentrating flow-through fractions</td>
</tr>
<tr>
<td>5188-5249</td>
<td>Luer-Lok adapters, pack of 2</td>
<td>Allows attachment of Luer-Lok syringes to spin cartridge</td>
</tr>
<tr>
<td>5188-5250</td>
<td>5-mL plastic Luer-Lok syringes, 1 pack of 2</td>
<td>For washing, eluting, and re-equilibrating buffers through spin cartridge</td>
</tr>
<tr>
<td>5188-5253</td>
<td>Needles, PTFE, Luer-Lock (1 pack of 10)</td>
<td>For transferring solutions with Luer-Lock syringes</td>
</tr>
</tbody>
</table>
Table 1  Agilent Multiple Affinity Removal Spin Cartridge and Accessories

<table>
<thead>
<tr>
<th>Agilent Part number</th>
<th>Product name</th>
<th>Description</th>
</tr>
</thead>
<tbody>
<tr>
<td>5188-5251</td>
<td>1.5-mL screwtop microtubes, 1 pack of 100</td>
<td>Eppendorf-style tubes used for collecting fractions from spin cartridge</td>
</tr>
<tr>
<td>5188-5252</td>
<td>Spin cartridge screw caps and plugs, 1 pack of 6 each</td>
<td>Extra caps and plugs for sealing the top and bottom of affinity spin cartridges</td>
</tr>
<tr>
<td>5188-5254</td>
<td>Starter Reagent Kit for Spin Cartridges</td>
<td>Buffer A: 1 L</td>
</tr>
<tr>
<td></td>
<td></td>
<td>Buffer B: 1 L</td>
</tr>
<tr>
<td></td>
<td></td>
<td>Spin filters 0.22 µm: 2 packs of 25</td>
</tr>
<tr>
<td></td>
<td></td>
<td>Protein concentrators: 1 pack of 25</td>
</tr>
<tr>
<td></td>
<td></td>
<td>Luer-Lok adapters: 1 pack of 2</td>
</tr>
<tr>
<td></td>
<td></td>
<td>5-mL plastic Luer-Lok syringes: 1 pack of 2</td>
</tr>
<tr>
<td></td>
<td></td>
<td>1.5-mL microtubes: 6 packs of 100</td>
</tr>
<tr>
<td></td>
<td></td>
<td>Spin cartridge extra caps and plugs, 1 pack of 6 each</td>
</tr>
<tr>
<td></td>
<td></td>
<td>Needles, PTFE, Luer-Lock (1 pack of 10)</td>
</tr>
</tbody>
</table>

Table 2  Additional materials required from external suppliers

<table>
<thead>
<tr>
<th>Material</th>
<th>Supplier</th>
</tr>
</thead>
<tbody>
<tr>
<td>Microcentrifuge with adjustable centrifugal force (capable of spinning at 100 × g) and timer</td>
<td>Eppendorf Model 5415D or equivalent</td>
</tr>
<tr>
<td>50-mL polypropylene tubes, or similar vessels to hold small quantities of Buffers A and B during procedure</td>
<td>General laboratory supplier</td>
</tr>
<tr>
<td>Adjustable pipettes for delivering up to 400 µL</td>
<td>General laboratory supplier</td>
</tr>
<tr>
<td>Transfer pipettes</td>
<td>General laboratory supplier</td>
</tr>
</tbody>
</table>

Storage Conditions

Upon receipt and when not in use, store the spin cartridge at 2°C to 8°C (35°F to 46°F). Store wetted with Buffer A and with the end-caps tightly sealed. **Do not freeze the spin cartridge.**
Overview

The Agilent Multiple Affinity Removal System comprises a family of immunodepletion products based on affinity interactions and optimized buffers for sample loading, washing, eluting, and regenerating. This spin cartridge is specifically designed to remove seven high-abundance proteins from human plasma samples. This technology enables removal of albumin, IgG, IgA, transferrin, haptoglobin, antitrypsin, and fibrinogen with a single device. The targeted high-abundance proteins are simultaneously removed when crude biological samples are passed through the cartridge. Selective immunodepletion provides an enriched pool of low-abundance proteins for downstream proteomics analysis, as depicted in Figure 1 on page 9.

Specific removal of the seven high-abundance proteins depletes approximately 88–92% of total protein mass from human plasma, facilitating study of the low-abundance proteins in the flow-through fractions. Removal of high-abundance proteins enables improved resolution and dynamic range for one-dimensional gel electrophoresis (1DGE), two-dimensional gel electrophoresis (2DGE) and liquid chromatography/mass spectrometry (LC/MS). The collected flow-through fractions may need to be concentrated dependent upon the downstream applications.

![Crude human plasma](image1)

![Low-abundance proteins free from interferences](image2)

![High-abundance proteins (albumin, IgG, IgA, transferrin, haptoglobin, antitrypsin, fibrinogen)](image3)

![Low-abundance proteins](image4)

**Figure 1**   The Multiple Affinity Removal System.
2

Instructions

Protocol for 0.45-mL spin cartridge
  Step 1. Prepare the materials
  Step 2. Prepare the sample
  Step 3. Prepare the spin cartridge
  Step 4. Load and run the cartridge
  Step 5. Analyze the flow-through fraction

Recommendations
Troubleshooting
Protocol for 0.45-mL spin cartridge

Step 1. Prepare the materials

During use, never let the spin cartridge frits or resin bed run dry. If this happens, see “Recommendations” on page 15 for a cartridge rewetting procedure.

1. Remove the Multiple Affinity Removal Spin Cartridge from cold storage and allow cartridge to equilibrate to room temperature before use.

2. Fill two 50-mL vessels with appropriate amounts of Buffers A and B, according to the number of samples being processed. The affinity removal protocol requires approximately 5 mL Buffer A and 2 mL Buffer B for each 12-µL plasma sample.

3. Label two 5-mL Luer-Lock syringes with “A” (for use on page 12 and page 13) and “B” (for use on page 13).

Step 2. Prepare the sample

Before you begin, consult the Certificate of Analysis for your cartridge to verify the cartridge capacity. Instructions below are based on a cartridge plasma capacity of 12–14 µL.

1. Dilute the 12 to 14-µL plasma sample with Buffer A to a final volume of 200 µL.

   If you plan to perform several successive runs on the cartridge, increase amount of diluted sample prepared in this step accordingly.

   Addition of protease inhibitors in Buffer A for sample dilution helps prevent protein degradation.

   The protocol may be applied to other human biological fluids like serum and CSF with necessary adjustments in sample volume based on albumin concentration.

2. To prevent clogging of spin cartridge frits, remove particulates with a 0.22 µm spin filter, spinning the diluted sample for 1 min at 16,000 × g.
Step 3. Prepare the spin cartridge

1. Remove cartridge cap and plug, retaining both for later cartridge storage.

2. Attach Luer-Lok adapter to spin cartridge.

3. Draw 4 mL of Buffer A into the syringe labeled “A” and then attach the syringe to the Luer-Lok adapter on the spin cartridge.

4. Dispense Buffer A slowly through spin cartridge to prepare resin and to remove any trapped air bubbles. With a transfer pipette, remove any excess Buffer A from top of the spin cartridge.

To avoid displacing cartridge frits, never apply negative pressure (e.g. by pulling up on the syringe plunger) or excessive positive pressure while dispensing solutions onto the cartridge.

If you experience high backpressure while loading any solution onto the cartridge, it is important to continue loading the solution slowly and to avoid applying extra pressure. See “Troubleshooting” on page 17 for additional suggestions.

Step 4. Load and run the cartridge

1. Remove the Luer-Lok adapter and place spin cartridge in a screw-top collection tube labeled F1 (for flow-through 1).

2. Add 200 µL of diluted plasma sample to top of resin bed.

3. Centrifuge for 1.5 minutes at 100 × g (or lowest possible setting on centrifuge - see Note below). Cap the spin cartridge loosely or leave open during centrifugation so the sample is able to flow. Collect the flow-through fraction in the collection tube F1. The resin bed and frits should remain moist, not dry, after centrifugation.

If centrifuge cannot be programmed to 100 × g, then cartridge capacity may be different for use; optimum depletion results are obtained when the flow rate is controlled to 0.2 mL/min.

4. Wash the cartridge with Buffer A and collect additional F1 flow-through fraction. To do this step, add 400 µL of Buffer A to top of resin bed and centrifuge for 2.5 minutes at 100 × g. Collect the flow-through into the same F1 collection tube used in step 3.
5  Do second wash with Buffer A and collect F2 flow-through fraction. To do this step, transfer spin cartridge to a fresh screw-top collection tube labeled F2, and add 400 µL of Buffer A to top of resin bed. Centrifuge for 2.5 minutes at 100 × g, collecting the flow-through into the F2 collection tube.

6  Remove the spin cartridge from the F2 collection tube and attach a Luer-Lok adapter to the cartridge top.

7  Elute the bound fraction from the cartridge with Buffer B. To do this step, fill the 5-mL Luer-Lok syringe labeled “B” with 2 mL of Buffer B and attach to the spin cartridge via the Luer-Lock adapter. Elute bound high-abundance proteins into a fresh collection tube by slowly pushing Buffer B through the spin cartridge. Save the bound fraction for analysis if desired, or discard it. Do not push air through the spin cartridge and do not allow the resin bed or frits to run dry.

   **NOTE**

   If the meniscus of Buffer B does not reach the top frit after depressing the syringe plunger completely, remove syringe and draw plunger back to the 1-mL mark with air and reattach to the cartridge. Use the air in the syringe as positive pressure to push Buffer B through until the meniscus of Buffer B reaches the top frit.

8  Re-equilibrate the cartridge with Buffer A. To do this step, fill the 5-mL syringe labeled “A” with 4 mL of Buffer A. Remove the Buffer B syringe from the cartridge and attach the Buffer A syringe. Slowly push Buffer A through the resin bed. Do not allow the resin bed or frits to run dry by leaving a small aliquot of buffer on the top of the frit.

9  After equilibration with Buffer A, the spin cartridge is ready for processing the next sample.

   If the spin cartridge will be stored before processing the next sample, leave the resin bed wetted with Buffer A, and leave a layer of Buffer A above the top frit. Re-seal both ends of the spin cartridge tightly, taking care not to displace or puncture the lower frit while placing plug in the lower end. Store the cartridge with ends capped at 2°C to 8°C (35°F to 46°F). **Do not freeze the cartridge.**
Step 5. Analyze the flow-through fraction

1. For maximum recovery of the low-abundance proteins, combine flow-through fractions F1 and F2. Alternatively, fractions F1 and F2 may be analyzed separately.

2. Analyze the flow-through fraction(s), containing the low-abundance proteins, to verify removal of the targeted high-abundance proteins using the guidelines below:
   - For 1D-SDS-PAGE, an aliquot of the flow-through fraction may be used directly.
   - For IEF, 2D-GE, and MS analysis of the flow-through fraction, it is necessary to do buffer exchange or desalt to an appropriate buffer. The 5 kDa MWCO spin concentrators (part number 5185-5991) may be used for buffer exchange and concentration. Alternatively, the Agilent mRP-C18 column (part number 5188-5231) may be used for automated desalting and concentration.
Recommendations

- **Sample dilution using Buffer A**
  Do not load crude plasma or other biological samples directly onto the cartridge. Follow instructions for plasma dilution with Buffer A on page 11.

- **Preventing protein degradation**
  Addition of protease inhibitors to Buffer A for sample dilution helps prevent protein degradation.

- **Sample cleanup**
  Human plasma may contain particulate materials that can be removed by a quick spin using a 0.22-µm spin filter.

- **Variation in cartridge capacity for different samples**
  Concentrations of the proteins targeted for depletion can vary among individual plasma samples and in different types of biological samples. Thus cartridge capacity for samples may differ and you may need to adjust the loading volume for a particular sample.

- **Spin Cartridge performance**
  Agilent Multiple Affinity Removal Spin Cartridges should perform reproducibly for greater than 200 runs when handled using the recommended procedures. Buffers A and B are optimized to support cartridge performance and longevity. We cannot guarantee cartridge performance if other buffers are used.

  Do not expose cartridges to organic solvents (like alcohols, acetonitrile, etc.), strong oxidizers, acids, reducing agents, or other protein-denaturing agents.

- **Cartridge rewetting**
  If the resin bed becomes dry during cartridge centrifugation or syringe elution, attach a syringe with Buffer A to the spin cartridge using a Luer-Lok adapter and re-equilibrate the cartridge by passing Buffer A through it. This should not affect the spin cartridge performance.

- **Spin Cartridge storage**
  To minimize loss in capacity, equilibrate the cartridge with Buffer A, cap the ends and store at 2°C to 8°C (35°F to 46°F). **Do not freeze the cartridge.**
• **Analysis of flow-through fractions**
  Buffer exchange to an appropriate buffer is recommended for high salt-sensitive applications such as IEF or MS. For 1D-SDS-PAGE, you can load flow-through fractions in Buffer A directly.

• **Concentration of flow-through fraction for further analysis**
  For further downstream proteomic analysis (e.g. SDS-PAGE or LC/MS), combine flow-through fractions F1 and F2 and concentrate the samples. Spin concentrators with 5 kDa MWCO (part number 5185-5991) can be used to concentrate proteins before analysis. Alternatively, the Agilent mRP-C18 column (part number 5188-5231) may be used for automated desalting and concentration.

• **Lyophilization of flow-through fraction**
  If lyophilization of the flow-through fraction (containing the low abundance proteins) is required after recovery from the cartridge, first do buffer exchange to a volatile buffer (such as ammonium bicarbonate). This is recommended due to the high salt concentration of the Buffer A solvent in the flow-through fraction.

• **Bound fraction analysis**
  If you wish to analyze the bound fraction, first do buffer exchange to phosphate-buffered saline (PBS) or to another buffer compatible with your analysis. Buffer B contains compounds that may interfere with some protein assays.
**Troubleshooting**

Review the following information for troubleshooting your experiments.

**Table 3  Troubleshooting suggestions**

<table>
<thead>
<tr>
<th>Problem</th>
<th>Cause</th>
<th>Solution</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>No flow</strong></td>
<td>Spin cartridge may be capped too tightly during centrifugation</td>
<td>Remove or loosen cap during centrifugation.</td>
</tr>
<tr>
<td>Bubble under resin or frits</td>
<td></td>
<td>Rewet with Buffer A (see the Cartridge rewetting section in “Recommendations”).</td>
</tr>
<tr>
<td><strong>Incomplete flow</strong> (also see No flow solutions, above)</td>
<td>Centrifugation parameters used do not produce the optimal flow rate</td>
<td>Adjust centrifuge force and time to achieve ≤0.2 mL/min flow rate through spin cartridge during sample loading on page 12 through Buffer A washes on page 13.</td>
</tr>
<tr>
<td><strong>Backpressure when loading solutions on cartridge</strong></td>
<td>Particulate matter is blocking frits</td>
<td>Filter samples prior to loading. Modify sample preparation methods to reduce amounts of hydrophobic compounds in sample.</td>
</tr>
<tr>
<td><strong>No proteins in bound fraction</strong></td>
<td>Buffers A and B reversed</td>
<td>Re-equilibrate spin cartridge with Buffer A (see step 8 on page 13) and start over, using correct buffer sequence.</td>
</tr>
<tr>
<td><strong>Breakthrough of high-abundance proteins in flow-through fractions F1 and F2</strong></td>
<td>Cartridge plasma capacity exceeded</td>
<td>Reduce plasma load per sample.</td>
</tr>
<tr>
<td></td>
<td>Plasma protein levels may be unusually high</td>
<td>Reduce plasma load per sample.</td>
</tr>
<tr>
<td></td>
<td>Flow rate through cartridge during sample loading too high</td>
<td>Reduce centrifugation force and/or time during sample loading to not exceed 0.2 mL/min flow rate.</td>
</tr>
</tbody>
</table>
This chapter contains reference information.
# Spin Cartridge Specifications

## Table 4  Specifications for Part Number 5188-6408

<table>
<thead>
<tr>
<th>Parameter</th>
<th>Description</th>
</tr>
</thead>
<tbody>
<tr>
<td>Type</td>
<td>Affinity depletion spin cartridge</td>
</tr>
<tr>
<td>Size</td>
<td>0.45 mL</td>
</tr>
<tr>
<td>Cartridge capacity</td>
<td>12 to 14 µL human plasma; consult the cartridge Certificate of Analysis.</td>
</tr>
<tr>
<td>Cartridge body material</td>
<td>Polypropylene</td>
</tr>
<tr>
<td>Frit material</td>
<td>Polyethylene with 35-µm pore size</td>
</tr>
<tr>
<td>Recommended centrifugal force</td>
<td>100 × g</td>
</tr>
<tr>
<td>Operating temperature</td>
<td>18–25 °C</td>
</tr>
<tr>
<td>Cartridge packing material</td>
<td>Affinity ligand-modified resin</td>
</tr>
<tr>
<td>Immobilized ligands</td>
<td>Affinity ligands to human albumin, IgG, IgA, transferrin, haptoglobin, antitrypsin, fibrinogen</td>
</tr>
<tr>
<td>Shipping solution</td>
<td>Buffer A with 0.02% sodium azide</td>
</tr>
<tr>
<td>Shipping temperature</td>
<td>2–8 °C (35–46 °F)</td>
</tr>
<tr>
<td>Storage temperature</td>
<td>2–8 °C (35–46 °F)</td>
</tr>
</tbody>
</table>
Related Agilent Products

Agilent Multiple Affinity Removal System spin cartridges and LC columns are listed in Table 5 below.

<table>
<thead>
<tr>
<th>Product Group</th>
<th>Proteins Removed</th>
<th>Format</th>
<th>Capacity</th>
<th>Part No.</th>
</tr>
</thead>
<tbody>
<tr>
<td>Human-14</td>
<td>albumin, IgG, IgA, transferrin, haptoglobin, antitrypsin, fibrinogen, alpha2-macroglobulin, alpha1-acid glycoprotein, IgM, apolipoprotein AI, apolipoprotein AII, complement C3, transthyretin</td>
<td>spin cartridge</td>
<td>8–10 µL plasma</td>
<td>5188-6560</td>
</tr>
<tr>
<td></td>
<td></td>
<td>4.6 x 50 mm LC column</td>
<td>up to 20 µL plasma</td>
<td>5188-6557</td>
</tr>
<tr>
<td></td>
<td></td>
<td>4.6 x 100 mm LC column</td>
<td>up to 40 µL plasma</td>
<td>5188-6558</td>
</tr>
<tr>
<td></td>
<td></td>
<td>10 x 100 mm LC column</td>
<td>up to 250 µL plasma</td>
<td>5188-6559</td>
</tr>
<tr>
<td>Human-7</td>
<td>albumin, IgG, IgA, transferrin, haptoglobin, antitrypsin, fibrinogen</td>
<td>spin cartridge</td>
<td>12–14 µL plasma</td>
<td>5188-6408</td>
</tr>
<tr>
<td></td>
<td></td>
<td>4.6 x 50 mm LC column</td>
<td>30–35 µL plasma</td>
<td>5188-6409</td>
</tr>
<tr>
<td></td>
<td></td>
<td>4.6 x 100 mm LC column</td>
<td>60–70 µL plasma</td>
<td>5188-6410</td>
</tr>
<tr>
<td></td>
<td></td>
<td>10 x 100 mm LC column</td>
<td>250–300 µL plasma</td>
<td>5188-6411</td>
</tr>
<tr>
<td>Human-6HC</td>
<td>albumin, IgG, IgA, transferrin, haptoglobin, antitrypsin</td>
<td>spin cartridge</td>
<td>14–16 µL serum</td>
<td>5188-5341</td>
</tr>
<tr>
<td></td>
<td></td>
<td>4.6 x 50 mm LC column</td>
<td>30–40 µL serum</td>
<td>5188-5332</td>
</tr>
<tr>
<td></td>
<td></td>
<td>4.6 x 100 mm LC column</td>
<td>60–80 µL serum</td>
<td>5188-5333</td>
</tr>
<tr>
<td></td>
<td></td>
<td>10 x 100 mm LC column</td>
<td>up to 340 µL serum</td>
<td>5188-5336</td>
</tr>
<tr>
<td>Human-6</td>
<td>albumin, IgG, IgA, transferrin, haptoglobin, antitrypsin</td>
<td>spin cartridge</td>
<td>7–10 µL serum</td>
<td>5188-5230</td>
</tr>
<tr>
<td></td>
<td></td>
<td>4.6 x 50 mm LC column</td>
<td>15–20 µL serum</td>
<td>5185-5984</td>
</tr>
<tr>
<td></td>
<td></td>
<td>4.6 x 100 mm LC column</td>
<td>30–40 µL serum</td>
<td>5185-5985</td>
</tr>
<tr>
<td>Human-HSA/IgG</td>
<td>albumin, IgG</td>
<td>spin cartridge</td>
<td>up to 50 µL serum</td>
<td>5188-8825</td>
</tr>
<tr>
<td></td>
<td></td>
<td>4.6 x 50 mm LC column</td>
<td>up to 100 µL serum</td>
<td>5188-8826</td>
</tr>
<tr>
<td>Human-HSA</td>
<td>albumin</td>
<td>spin cartridge</td>
<td>up to 75 µL serum</td>
<td>5188-5334</td>
</tr>
<tr>
<td></td>
<td></td>
<td>4.6 x 50 mm LC column</td>
<td>up to 175 µL serum</td>
<td>5188-6562</td>
</tr>
<tr>
<td>Mouse-3</td>
<td>albumin, IgG, transferrin</td>
<td>spin cartridge</td>
<td>25–30 µL serum</td>
<td>5188-5289</td>
</tr>
<tr>
<td></td>
<td></td>
<td>4.6 x 50 mm LC column</td>
<td>37–50 µL serum</td>
<td>5188-5217</td>
</tr>
<tr>
<td></td>
<td></td>
<td>4.6 x 100 mm LC column</td>
<td>75–100 µL serum</td>
<td>5188-5218</td>
</tr>
</tbody>
</table>
Additional related products for use with the Agilent Multiple Affinity Removal System are listed in Table 6 below.

<table>
<thead>
<tr>
<th>Part Number</th>
<th>Description</th>
<th>Content</th>
</tr>
</thead>
</table>
| 5185-5986   | Starter Reagent Kit for Multiple Affinity Removal System LC columns | Buffer A: 2 x 1 L  
Buffer B: 1 L  
Spin filters 0.22 µm: 2 packs of 25  
Protein concentrators: 1 pack of 25 |
| 5188-5254   | Starter Reagent Kit for Multiple Affinity Removal System spin cartridges | Buffer A: 1 L  
Buffer B: 1 L  
Spin filters 0.22 µm: 2 packs of 25  
Protein concentrators: 1 pack of 25  
Luer-Lok adapters: 1 pack of 2  
5-mL plastic Luer-Lok syringes: 1 pack of 1  
1.5-mL microtubes: 6 packs of 100  
Spin cartridge extra caps and plugs, 1 pack of 6 each  
Needles, PTFE, Luer-Lock (1 pack of 10) |
| 5188-5231   | mRP-C18 High Recovery Protein Fractionation and Desalting Column | 1 Column  
(see www.agilent.com for product details) |
In This Book

This document describes how to use the Multiple Affinity Removal Spin Cartridge Human-7, 0.45 ml to chromatographically remove interfering high-abundance proteins from human biological samples.