Agilent Captiva EMR—Lipid Method Guide

General instructions for 96-well plate and 1 mL cartridge formats:

Agilent Captiva EMR—Lipid cartridges and 96-well plates allow streamlined in-well protein precipitation, filtration, and cleanup for lipid-containing samples. The improved filter design gives easy flow with vacuum or positive pressure, and allows for protein precipitation without clogging during elution. The novel EMR—Lipid sorbent chemistry provides highly selective and efficient lipid/matrix removal without impacting analyte recovery. Effective matrix removal assures minimal ion suppression or enhancement on target analytes, which significantly improves method reliability and ruggedness.

The 96-well plates are ideal for high-throughput workflows, while 1 mL cartridges can accommodate small batch needs.

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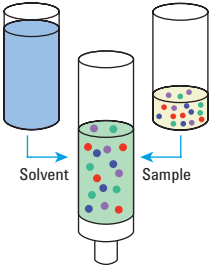
Operating instructions and tips for Agilent Captiva EMR—Lipid 96-Well Plate and 1 mL Cartridge Products

1. Add biological sample and crash solvent

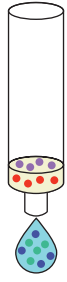


2. Mix to precipitate protein

Option 1 – **Passive mixing:** let stand for 5 minutes

Option 2 – **Active in-well mixing:** vortex or mix with wide-bore pipette tips

3. Filter



It is highly recommended to add sample first and then crash solvent, to achieve better sample homogeneity during sample and solvent addition.

● Salts ● Proteins ● Lipids ● Analyte

User Tips

Protein precipitation workflow	
Product configuration	96-well plate or 1 mL cartridge
Sample size	Between 20–200 µL
Sample treatment	Crash solvent ratio: between 3:1 and 5:1 ACN with 1 % formic acid to sample. If total volume is less than 500 µL, add additional 4:1 ACN:H_2O to reach a minimum volume of 500 µL. ACN is preferable to MeOH to maximize protein precipitation and avoid gelation.
Recommended sample addition order	1) Sample 2) Crash solvent
Mixing	**Option 1:** Passive mixing. Let stand for 5 minutes to allow for complete protein precipitation to occur. **Option 2:** Active in-well mixing. For 96-well plates containing >500 µL of total volume and 1 mL cartridges regardless of volume, pipette mixing using wide bore pipette tips (perform 3 to 5 aspiration/dispense cycles) is recommended. For 96-well plates with <500 µL of total volume, cap and vortex at 1,350 rpm. **Option 3:** Protein precipitation and mixing can be performed in a separate tube, centrifuged, and subsequently transferred to the Captiva EMR—Lipid well/cartridge.
Pass-through filtration and cleanup	Vacuum between 2–5 in Hg initiates flow. Positive pressure (3–4 psi) is also acceptable. **For optimal lipid removal, a controlled flow rate of one drop every 3–5 seconds is highly recommended.** After elution, apply higher vacuum or positive pressure for 1–2 minutes to ensure maximum sample recovery. Flow rate is dependent on sample type, crash solvent, and mixing. An alternative approach to vacuum and positive pressure is centrifugation. For 96-well plates, 500–800 rpm for a minimum of 10 minutes is recommended, followed by 1–2 minutes at 2,500–3,000 rpm to drain the cartridge. Centrifugation speed and time are dependent on the sample volume and matrix.

For more detailed protocol recommendations, please refer to Agilent Application Note 5991-9222EN.

Agilent Captiva EMR—Lipid ordering information

Part number	Description	Quantity
5190-1000	Agilent Captiva EMR—Lipid 96-well plate	1 plate
5190-1001	Agilent Captiva EMR—Lipid 96-well plate	5 plates
5190-1002	Agilent Captiva EMR—Lipid 1 mL cartridge	100/pk

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