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Lipid Extraction for Mass Spectrometry Lipidomics

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We use this protocol and it's working

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Abstract

This protocol details how to perform lipid extraction using a methyl tert-butyl ether (MTBE)-based extraction method compatible with mass spectrometry.

Lipid Extraction

1 Add 200 µL of cold methanol and 800 µL of cold methyl tert-butyl ether (MTBE) to the samples.

2 Vortex.

3 Add 200 µL of water. Phase separation into aqueous and organic phases should occur.

4 Centrifuge the extraction mixture at 10000 x g, 4°C, 00:10:00 to separate the organic and the aqueous phases. 10m

5 Collect the upper organic phase.

6 Dry it in a SpeedVac and store at -80 °C until analysis.

7 For mass spectrometry analysis, reconstitute the samples with 20 µL - 40 µL of acetonitrile/isopropanol/water in a 65:30:5 ratio (v/v/v).

> Note
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> For LC-MS measurement, a C8 column (Luna 3u, 2 × 100 mm, 3.0 µm, Phenomenex) is recommended. DMPC gets stuck in the C18 column.