Each sample was extracted using a modified Bligh-Dyer extraction.
Briefly, filters were cut up and split between two bead beating tubes containing a mixture of 100 µm and 400 µm silica beads.
Heavy isotope-labeled internal standards were added along with 750 µL of cold aqueous solved (50:50 methanol:water) and 750 µL of cold organic solvent (dichloromethane).
The samples were shaken on a FastPrep-24 Homogenizer for 30 seconds and chilled in a -20 ∞C freezer repeatedly for three cycles of bead-beating and a total of 30 minutes of chilling.
The organic and aqueous layers were separated by spinning samples in a microcentrifuge at 5,000 rpm for 90 seconds at 4 ∞C.
The aqueous layer was removed to a new glass centrifuge tube.
The remaining organic fraction was rinsed three more times with additions of 750 µL of 50:50 methanol:water.
All aqueous rinses were combined for each sample and dried down under N2 gas.
The remaining organic layer was transferred into a clean glass centrifuge tube and the remaining bead beating tube was rinsed two more times with cold organic solvent.
The combined organic rinses were centrifuged, transferred to a new tube, and dried under N2 gas.
Dried aqueous fractions were re-dissolved in 380 µL of water.
Dried organic fractions were re-dissolved in 380 µL of 1:1 water:acetonitrile.
20 µL of isotope-labeled injection standards in water were added to both fractions.
Blank filters were extracted alongside samples as methodological blanks.