LC-MS/MS analysis

The hydrophobic and hydrophilic metabolites from each extracted sample were resolved by reverse-phase chromatography (RPLC) using a Syncronis C18 column (150 x 4 mm x 5 µm, Thermo Fisher Scientific) and hydrophilic interaction liquid chromatography (HILIC) using a Hypersil Gold HILIC column (100 x 2.1 mm x 3 µm, Thermo Fisher Scientific), respectively. Specific mobile phase compositions were employed for each separation mode: For RPLC, Mobile phase A contained 0.1% formic acid in water, while Mobile phase B contained 0.1% formic acid in methanol. For HILIC: Mobile phase A contained 95:05 acetonitrile and water, while Mobile phase B contained 30:70 acetonitrile and water. Following chromatographic separation, the isolated metabolites were analysed using an Agilent 6550 iFunnel Q-TOF LC/MS system with electrospray ionization (ESI) (Agilent Technologies). Both positive and negative ionization modes were used to acquire full MS scan data. Full MS scans were acquired from m/z 60 to 1000 at a scan rate of 1 spectra/sec. Following separation by LC, individual chemical peaks within the sample were identified using MS/MS fragmentation and subsequent deconvolution with Agilent MassHunterTM Qualitative Analysis B.06.00 (MassHunterTM Qual, Agilent Technologies).