Orbitrap Q Exactive Plus

Positive ionisation: instrument is externally calibrated using Caffeine, MRFA (MET-ARGPHE-ALA), and Ultramark 1621.

Negative ionisation: instrument is externally calibrated using Caffeine, MRFA, Ultramark 1621, Sodium Dodecyl Sulfate and Sodium Taurocholate every week.

The Orbitrap Q Exactive Plus also set to use internal calibration with three ions (Lockmass) during accurate mass measurement analyses.

Loop-injection method a Vanquish LC system is used to provide solvent flow at 300 µL/min., through a 10-µL sample loop into which the sample is injected. Around 5 µL of sample diluted in 1 mL of methanol and 2 µL of the resulting sample injected onto a C_4 column, 1.9 µm pore size, 2.1 mm x 150 mm (Thermo Scientific, UK). Mobile phases are (A) water 0.1% formic acid (B) acetonitrile, 0.1% formic acid. The gradient is as follows: 5% B for 1 min with a linear increase to 95% B over 4 min, in 0.1 min decrease to 5% B and for 0.9 min stays at 5% B. The LC-MS analysis time is 6 min.

API source settings HESI ion source temperature between 200°C to 350°C, Sheath Gas flow 10 to 30 (arb. units), Aux Gas flow 5 to 15 (arb. units), spray voltage positive ionisation: + 3.2 to 3.7kV negative ionisation: - 3.5 to - 4.0kV, Probe heater temperature: 100 to 320 °C these parameters would depend on samples analysed.

Mass spectra acquired at a minimum resolution of 30,000 (at *m/z* 400).

Specifications as per ThermoScientific

- Resolution 7,500, 15,000, 30,000, 60,000 or 100,000 (FWHM) @ *m/z* 400 with a scan repetition rate of 1 second
- Mass Range *m/z* 50–2000; *m/z* 200–4,000
- Mass Accuracy <3 ppm RMS for 2 h period with external calibration using defined conditions, <2 ppm RMS with internal calibration
- Dynamic Range >10,000 between mass spectra, >4,000 between highest and lowest detectable mass in one spectrum