

## AbsoluteIDQ® p400 HR kit requirements

This document specifies additional lab equipment and chemicals that are required but not included with the kit. Before starting with the kit, ensure that all required items are available.

### LC-MS system

Thermo Scientific	
Mass spectrometer with <u>HESI-II</u> ion source	
	<ul style="list-style-type: none"> <li>– Q Exactive™</li> <li>– Q Exactive™ Focus</li> <li>– Q Exactive™ Plus</li> <li>– Q Exactive™ HF</li> </ul>
LC and autosampler	
	<ul style="list-style-type: none"> <li>– Standard flow UHPLC system with column oven. NanoLC and microLC are not supported!</li> <li>– 96-well plate autosampler with temperature control (10 °C)</li> <li>– Injection volume range: 5 – 20 µL</li> <li>– Special sample rack for deep-well plates required for the following autosamplers:               <ul style="list-style-type: none"> <li>– UltiMate 3000: Thermo part no. 6820.4083 (30 – 36 mm height)</li> </ul> </li> </ul>
Tubing	
	<ul style="list-style-type: none"> <li>– Required between injector and column system: Thermo Scientific™ Viper™ Capillary 0.13 x 350 mm (6040.2335)</li> <li>– Recommended between column and mass spectrometer: Red PEEK Tubing 1/16" OD x 0.005" ID</li> </ul>

### LC-MS instrument status

Condition	
	<ul style="list-style-type: none"> <li>– <b>Serviced, calibrated and tuned instruments</b> according to manufacturer's recommendations on a regular basis</li> <li>– LC-MS system must be <b>free of ion-pair reagent</b></li> </ul>

### Analytical column

UHPLC	
Analytical column	AbsoluteIDQ® p400 HR kit UHPLC column: biocrates product number 9120052121032

## Solvents and chemicals

Solvents and chemicals	Purity
Acetonitrile, ethanol, isopropanol, methanol, water	LC-MS grade (ethanol: HPLC grade sufficient)
Formic acid	LC-MS grade (e.g. Honeywell Fluka™ 56302-50ML) <b>Fresh! New bottle or opened within past 6 months</b>
Phenyl isothiocyanate (PITC)	99%, for protein sequencing (e.g. Sigma-Aldrich 317861) <b>Fresh! New bottle or opened within past 6 months</b>
Pyridine	Purity >99% (recommended Acros Organics 131780500, 50mL) <b>Fresh! New bottle or opened within past 6 months</b>
Ammonium acetate	LC-MS grade
Phosphate buffered saline (PBS)	p.a. grade (e.g. Sigma-Aldrich P4417)

## Laboratory equipment

Laboratory equipment		
	Nitrogen evaporator for 96-well plates	Pressure manifold for 96-well plates
Nitrogen evaporator* <u>or</u> pressure manifold* > <b>Requires nitrogen supply</b> > <b>Must be in a fume hood</b>	<b>Examples:</b> – Porvair blowdown evaporator MiniVap® – Techne (FSC496D) – Sample concentrator from BenchTop Lab Systems (96 samples, BT1604) – VLM evaporators – Organomation MICROVAP microplate evaporator – Biotage TurboVap® 96	<b>Examples:</b> – Waters positive pressure-96 processor (186006961) – Biotage® PRESSURE+ 96 manifold (PPM-96) – CEREX® system 96 processor (288-0001) – Agilent positive pressure manifold 96 processor (PPM-96) – TECAN Resolvex® M10 96
Centrifuge	Must be able to centrifuge 96-well plates of 5 cm height at 500 x g	Not required when a pressure manifold is used.
Shaker	Any model with adjustable speed (450 – 1200 rpm) including a tray for plates and vials. Recommended: Eppendorf MixMate® or ThermoMixer®	
Pipettes	– Repeater, e.g. Eppendorf Multipette® E3 (with 1.0, 2.5 and 10 mL tips) or similar electronic model – Single channel: volume range 10 µL – 1000 µL – 8-Channel: 150 µL	
Vortexer	Any model	
Balance	Accuracy < 1 mg	
Solvent bottles	50 – 1000 mL	

\* See document “Technical guide-Nitrogen evaporators and pressure manifolds” for details.

## Software

Thermo Scientific
MS software
Xcalibur™ software 4.0 or later
Data processing computer
<ul style="list-style-type: none"><li>– Oracle® database:<ul style="list-style-type: none"><li>– <b>Option 1 (recommended):</b> full commercial version, updates and support provided by Oracle. Expert required.</li><li>– <b>Option 2:</b> Oracle Database Express Edition (Oracle XE), free of charge, provided by biocrates. No updates and support provided by Oracle.</li></ul></li><li>– MetIDQ™ software (provided by biocrates)</li><li>– Windows 7 or later, 64-bit architecture</li><li>– Microsoft .NET Framework 4.5.2 or later</li><li>– Microsoft Visual C++ redistributables (32 &amp; 64 bit each): 2005, 2008, 2010, 2012, and 2013</li></ul>