

Sample preparation for MetIDQ p180 Kit measurement

Solvents:

Acetonitril (Merck KGaA, Darmstadt, Germany hypergrade for LC-MS)

Water MiliQ,

Extracting agent - ACN / H₂O (1:1)

Equipment

4 steel balls size M

Eppendorf Tubes 2mL

Tissue slicer (Rettberg, Germany)

Centrifuge (Sigma)

Work steps

Approximately 100mg of caecum sample and 4 steel balls of size M into each tube. Per mg of sample 5µL of extracting agent was added. Shake the samples for 10 minutes at 30 Hz in the tissue slicer and centrifuge for 2 minutes at 14000 rpm. 10 µL supernatant was used for the targeted analytics.

Blood serum samples 10 µL were used for analysis

Kit reparation

The analysis was performed using the validated MetIDQ p180 Kit and described in Siskos et al. [1]. Data processing was carried out with the provided quantitation method Kit (Biocrates Life Sciences AG, Innsbruck, Austria).

1 Siskos AP, Jain P, Römisch-Margl W, Bennett M, Achaintre D, Asad Y, *et al.* Interlaboratory Reproducibility of a Targeted Metabolomics Platform for Analysis of Human Serum and Plasma. *Anal Chem* 2017;**89**:656-65.