Sample Preparation Protocol for ESI Accurate Mass Service

Only **novel samples that need accurate mass confirmation** and have already been run on Open Access systems should be submitted to this service.

Please aim for an analyte concentration of around **10 μg/mL.** When analyte concentration cannot be estimated, we may not be able to run your samples.

Over concentrated samples lead to increased chemical noise, poor mass resolution, blockage in the sample flow path and contamination of the mass spectrometer. You should be able to see through your sample vial even if the solution is coloured. Please also make sure that there are no hard particles in the solution or precipitation at the bottom of your sample vial, and that the solution is not jelly-like or cloudy.

Only standard **2 mL Mass Spec sample vials** with a **pre-slit septum** on the top of the lid should be used. The vials are available from CRL stores. No taller vials or vials with hard lids can be used.

All open access instruments use **electrospray ionisation** which is only **compatible with volatile organic solvents and water.** Samples must be cleaned of inorganic salts: **high inorganic salt concentrations are not compatible with ESI.** Please follow the protocol below for sample preparation.

The following approach is recommended for making up samples for **High Resolution ESI service:**

1. Dissolve the sample in any organic solvent (e.g., DCM, $CHCl_3$, EtOAc, MeCN, MeOH) or H_2O to a concentration of 1 mg/mL. Please ***do not use low vapour pressure solvents***, such as DMSO, or dilute them >20-fold in another solvent.
2. Take 10 uL of this solution and dilute it with 1 mL of either methanol, acetonitrile or water (or any combination of these solvents).
3. If there is any precipitate in the resulting solution ***it must be filtered*** before running the sample otherwise this is very likely to cause line blockages and delays with sample analysis for all users.
4. Place the solution in *a standard* ***2mL Mass Spec*** **vial** with a screw cap lid and **pre-slit septum** on the top (available from stores).
5. ***Do not use Trifluoroacetic acid (TFA) in your samples***. If you need to acidify your samples use formic acid.
6. ***Do not use Tetrabutyl ammonium (TBA) in your samples*** (also avoid other ion-pairing agents) these will contaminate all subsequent samples run on the system.