



SAMPLE PREPARATION GUIDE FOR SYNTHETIC ORGANIC CHEMICALS

Proper sample preparation is an important prerequisite for obtaining reliable results. Please follow the general guideline to prepare your sample for mass spectrometry analysis.

Before you come to BNRF:

- Try your best to remove solvent from your sample. The sample should usually be solvent-free and pure.
- Avoid or minimize the sample content of the following ingredients:
 - Salts in general, sodium and phosphate salts in particular. The salts from synthetic source, such as NaCl, Na₂SO₄, Na₂CO₃, or NaHCO₃, should be removed.
 - Viscous compounds (DMSO, glycerol).
- Understand the solubility of your sample in different solvents. We prefer to dissolve the sample in MeOH, ACN, and H₂O.
- 1 mg is always enough. Place your sample powder/oil in a clean glass bottle or eppendorf tube. Bring your solvent-free sample to BNRF and dissolve your sample with BNRF LC/MS grade solvents.
- Calculate the molecular weight of your sample. Fill the submission form and bring it with your sample.

Sample preparation at BNRF:

- Samples should be prepared in a water-soluble organic solvent or solvent/water mix and filtered via 0.2 um syringe filter or centrifuged. An acid or base to 0.1% v/v may also be added.
- Centrifugation of sample is highly recommended prior to analysis since small particulate matter can block the capillary systems of the instrument.
- Samples should be prepared to a concentration range 1-50µg/mL.

Notes:

- Ionisation can be enhanced by promoting protonation or deprotonation of the analyte. For example, addition of 0.1% acetic acid to an analyte

containing an amino functional group will protonate the amine, hence ($M+H^+$) will be detected in positive ion mode. Addition of 0.1% NH_4OH to an analyte containing an acidic functional group will deprotonate the acid, hence ($M-H^+$) will be detected in negative ion mode.

- Only volatile buffers should be used if needed to prepare analyte, i.e. ammonia, acetic or formic acid. Avoid the use of non-volatile buffers i.e. phosphates, borate

ESI compatible solvents:	<ul style="list-style-type: none"> • Acetonitrile (CH_3CN) • Dichloromethane (CH_2Cl_2) - if mixed with methanol • Dichloroethane (CH_2ClCH_2Cl) • Tetrahydrofuran (THF) • Ethanol (CH_3CH_2OH) • Propanol ($CH_3CH_2CH_2OH$) • Methanol (CH_3OH) • Nitromethane (CH_3NO_2) • Toluene ($C_6H_5CH_3$) - if mixed with methanol or acetonitrile • Water
Solvents tolerable in small amounts:	<ul style="list-style-type: none"> • Dimethylsulphoxide (DMSO) ($(CH_3)_2SO$) • Dimethylformamide (DMF) ($HCON(CH_3)_2$)
Solvent Modifiers / Additives Compatible with ESI:	<ul style="list-style-type: none"> • Volatile Salts or Buffers eg. ammonium acetate (NH_4CH_3COOH), ammonium bicarbonate (NH_4HCO_3) • Volatile Acids eg. formic acid ($HCOOH$), acetic acid (CH_3COOH)
Solvent Modifiers / Additives NOT Compatible with ESI: (tolerable in small amounts)	<ul style="list-style-type: none"> • Trifluoroacetic acid (TFA) • Heptafluorobutyric acid • Sodium Dodecyl Sulphate (SDS) • Ethylenediaminetetraacetic acid (EDTA) • Involatile Salts or Buffers eg. sodium chloride ($NaCl$), phosphates (NaH_2PO_4)

Reference:

1. <http://www.chem.ualberta.ca/~massspec/ESIsol.htm>
2. <http://webcache.googleusercontent.com/search?q=cache:9zYIVPidxxoJ:https://pharmacy.tcd.ie/assets/doc/Mass%2520Spec%2520requisition%2520form.doc+&cd=1&hl=en&ct=clnk&gl=us>