Liquid NMR sample

- **Keep holders and rotors clean**
- **Adjust solvent volume**

  ~4cm - 4.5cm (600ul) is recommended
- **Fit sample in gauge**

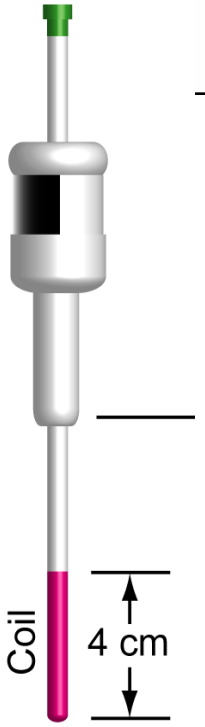
  If less than 4cm, adjust the sample centre to the gauge 0 Position

  Fit the O-ring inside the rotor to the holder, pull and rotate the holder against the rotor.
- **Make a homogeneous solution**

  Shake your sample

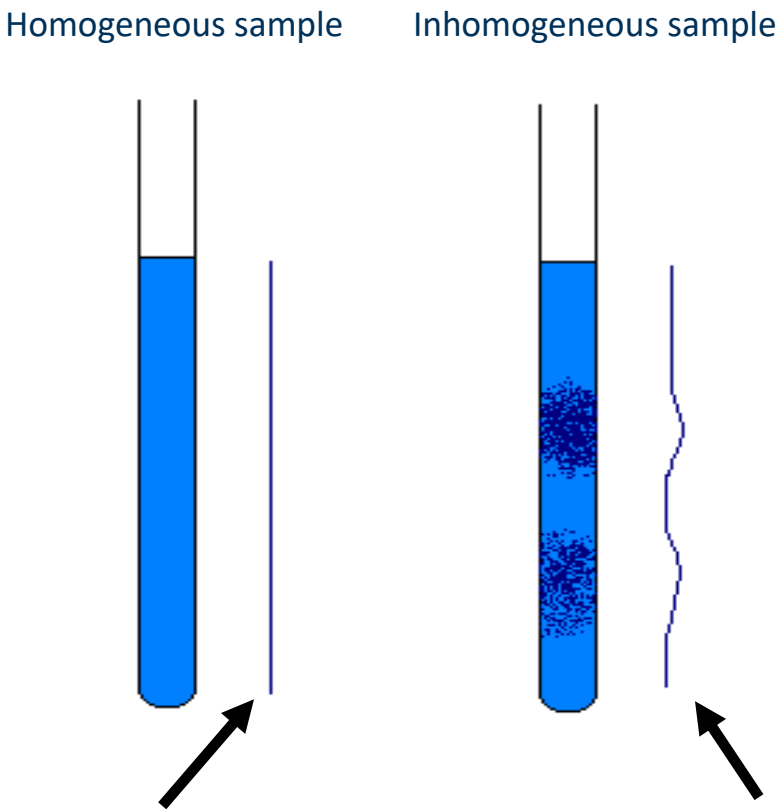
  Do not concentrate too much
- **Remove or filter insoluble material**

  Detection area needs to be clean



Innovation JEOL

Sample homogeneity



Magnetic field (magnetic susceptibility) is also homogeneous

Magnetic field (magnetic susceptibility) is also NOT homogeneous

If your compound is not homogeneously dissolved, it causes high order distortion which is impossible to adjust by shimming

Just shake your sample
But do not let bubbles stay around the detection area

Solutions for Innovation JEOL

Solvent choice

- Consider positions of solvent resonances
- Consider possibility of proton exchange
- Choose solvent with appropriate boiling or freezing point
- Select optimum concentration (based on available sample)
- Minimize solution viscosity (for high resolution)
- Consider possible reactivity
- Consider temperature dependence of solubility
- $CDCl_3$ often has acid (DCl), if your compound is sensitive to acid, be careful. Some vendors sell $CDCl_3$ with silver foil(as radical scavenger).
- Solvents affect chemical shift. For example acetone and benzene can displace chemical shifts significantly compared with chloroform.

Common NMR solvents

DOI: 10.1021/om100106e, 10.1021/jo971176v, 10.1021/acs.oprd.5b00417 & www.nmrs.io

Solvent	δ_H (ppm)	δ_{HDO} (ppm)	δ_c (ppm)	FP (°C)	MP (°C)
Acetone-d_6	2.05	2.0	206.7, 29.9	-94	57
Chloroform-d	7.27	1.5	77.2	-64	62
Deuterium oxide-d_2	4.80	4.8	---	4	101
Dichloromethane-d_2	5.32	1.5	54.0	-95	40
Dimethylformamide-d_7	8.03, 2.92, 2.75	3.5	163.2, 34.9, 29.8	-61	153
Dimethylsulfoxide-d_6	2.50	3.3	39.5	18	189
Methanol-d_4	4.87, 3.31	4.9	49.2	-98	65
Pyridine-d_5	8.74, 7.58, 7.22	5.0	150.4, 135.9, 123.9	-42	114
Tetrahydrofuran-d_8	3.58, 1.73	2.4	67.6, 25.4	-109	66
Toluene-d_8	7.09, 7.00, 6.98, 2.09	0.4	137.9, 129.2, 128.3, 125.5, 20.4	-95	111
Trifluoroacetic acid-d	11.30	11.5	164.2, 116.6	-44	75

Solutions for Innovation JEOL

Reference compounds

- Addition of chemical-shift standard (*e.g.*, TMS) is no longer essential because shift referencing is based on absolute frequencies automatically or use a solvent signal
- May use internal standard for quantitation
- May place reference compounds in capillary inserts

Solutions for Innovation JEOL

NMR tubes

- Why are tubes so expensive?
- Must meet strict standards of camber (straightness) and concentricity
- Lower-quality tubes may lead to artifacts (usually spinning sidebands), particularly in ^{1}H spectra
- “Disposable” tubes are generally of lower quality, but *may* be adequate for some purposes
- Try different vendors to find best value for the intended purpose
- Poor quality tubes may damage the probe

NMR tubes characteristics

- **Concentricity**: How centered is the internal diameter of the tube. Poor concentricity will lead to worse lineshapes and worse spinning sidebands when spinning. Higher frequency specified tubes typically have better concentricity to achieve better lineshapes.
- **Camber**: How straight is the tube. Poor camber will lead to worse lineshapes and worse spinning sidebands when spinning. Very poor camber will damage the probe. Higher frequency specified tubes typically have better camber to achieve better lineshapes.
- **Outside Diameter**: it needs to be set so it fits in the spinner without damaging it or slipping through it. Forcing a large tube into a spinner will damage o-rings of spinner and will likely damage the probe.
- **Inside Diameter**: This will depend on the wall thickness, but the tolerance will determine the precision of your external quantitation (if you are comparing integrals in the same spectrum this does not matter). For example, if you have a 1mM sample
  - in a tube specified as 4.2± 0.2mm, the effective concentration detected by NMR can vary by 9.75% in different tubes
  - in a tube specified as 4.2065 ± 0.0065 mm, the effective concentration detected by NMR can vary by 0.31% in different tubes
- **Temperature resistance**: Economy tubes cannot handle well temperature jumps. Do not use them for experiments with temperature variations. Do not reuse them as they may get bent if heated for cleaning/drying.

https://web.archive.org/web/20080513142827/http://www.wilmad-labglass.com/services/NMR_010.jsp
https://web.archive.org/web/20080827224550/http://www.wilmad-labglass.com/services/NMR_001.jsp

Tube tester

- Our current probes are built with very low tolerances in order to achieve the best specifications. The drawback of this is that any tube that is slightly bent will damage the glass insert in the probe when spun, and this is a repair that is not covered by warranty, because is misuse of the system. In order to verify that the tubes are straight, we can put the NMR tube in a tube tester and if it passes smoothly, then it is fine to be used in the probe.

https://www.wilmad-labglass.com/Products/SB-5-7/

Tube recommendations

- Dirt may also contribute to unstable spinning which can also crack the glass insert so cleanliness is essential at all times:
- Economy/high throughput tubes are not reusable. They state in their specifications that they cannot handle VT operations, and thus cleaning and drying them will bend them.
- Precision tubes can be reused but must be dried on a flat surface at low temperature to prevent glass distortion.
- Tubes and spinners must be cleaned before using them to prevent introducing any dirt in the probe (kimwipe or kimwipe slightly moist with IPA).
- Any tube scratched or damaged in any way must be disposed. Tubes with cracked glass at the top, unlevelled caps or with labels sticking out will be unbalanced and thus will not spin straight.
- If any ink is used to label the tubes, this must be permanent ink, if permanent ink is getting smeared around the tubes due to solvents it is equally bad and other ways of labelling must be used.
- Cleaning spinners with a lint-free wipe moist with IPA may be necessary every few months, depending on how quickly the dirt builds up (solvents may dissolve sticker used to detect spinner).

Cleaning of NMR tubes: https://www.wilmad-labglass.com/Support/NMR-and-EPR-Technical-Reports/Proper-Cleaning-Procedures-for-NMR-Sample-Tubes/

Tube type with mass limited samples

- Shigemi tubes or susceptibility plugs to concentrate all mass on the volume detected by the coil
  - Expensive/shimming difficulty/difficult to handle
- 3mm tubes (3mm spinners/adaptors to 5mm spinners/ 5mm with bottom part of the tube with reduced diameter/3mm tube with spacers inside 5mm tube)
  - No sensitivity advantage in use smaller diameter tube if filled to the same height

Volumes
4.5mmx40mm. Vol 6.36E-7 ~/1.14
4.2mmx40mm. Vol 5.54E-7
2.4mmx40mm Vol 1.81E-7 ~3
2.4mmx30mm Vol 1.36E-7 ~4
2.4mmx5mm Vol 2.3E-8 ~20

So we can in principle

sensitivities
3mm HX 1H 300
3mm TH 13C 100
5mm RO 1H 800/1.14 = 700
5mm RO 13C 270/1.14 = 236

In terms of official specs then
If we were to fill to the same height, the advantage of a 3mm probe would be
300*3/700 = 1.3
100*3/236 = 1.3

If we were to fill to the recommended height, the advantage of a 3mm probe would be
300*4/700 = 1.7
100*4/236 = 1.7

In practice, this advantage is higher

Dr Paululat 14.09 Day, 2021