

PREPARATION OF AN NMR SAMPLE

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You are about to use a procedure with expensive samples and where mistakes are not easily reversed. It requires careful analytical thought. Take your time, and read this document several times.

When you prepare an NMR sample, use the following guidelines, which are written for chloroform-d + TMS + your compound. You need a clean dry vial. If a drop of water or acetone remains in the vial, it will make a peak which is usually as large as your compound's peaks. You also need a clean cap for the vial. Do not attempt to wash a cap. Instead, select one which already looks clean. The reason for this is that cap liners absorb water or organics, and even a Teflon liner traps water or organics between the liner and the cap.

You need a clean dry Pasteur pipette. Do not attempt to wash out a rubber bulb. Tap it vigorously on a paper towel to make sure it is dry. You should never pull solutions into a rubber bulb because it is a source of multiple contaminants. Even a clean dry rubber bulb contains crosslinking compounds which can dissolve in a solvent.

A bulb which has been contaminated with a solid or high boiling liquid can be washed out. Make the final rinse acetone, and give it an extended drying period.

For solids, you need a clean dry spatula.

The reason for the clean dry items in the above paragraphs is that NMR solvents generally cost 100 to 1000 times as much as "normal" solvents. NMR solvents are deuterated, and normal solvents are not.

Do not grease any joints if you distill your compound. Silicone grease is very soluble in common NMR solvents and gives a peak near TMS. It can be impossible for a beginner to choose between them to set $\delta = 0$ ppm in an NMR tube. Hydrocarbon grease is also soluble, and gives hydrocarbon peaks. Make sure the joint at the top of the pot is clean after you pour in your compound. Wipe it dry if your compound doesn't evaporate. This greaseless procedure produces a frozen joint between the pot round bottom and distillation head if your unknown oxidizes or polymerizes in the joint during a high temperature distillation. Teflon sleeves without grease work for 1 atm distillations. Be careful about the ice on your receiver, leaking water from the hoses, and condensation in hot humid weather. Water is highly visible in a ^1H NMR.

Add 0.8 mL of chloroform-d/TMS to your empty vial.

Pipette 10 drops of the distilled liquid compound into the vial containing the NMR solvent. The pipetted solute should be at room temperature before pipetting. Because of oxidation over the several year lifetime of a reagent bottle, high boiling contaminants can constitute more than 0 % of the undistilled compound. For this reason, an undistilled liquid should never be used. Oxidation almost never produces compounds which boil below the compound, so the oxidized contaminants will remain in the pot if separation is good enough.

For solids, put 50 mg of the solid compound into the vial containing the NMR solvent.

Don't attempt to ice the solution to prevent TMS from evaporating. If you do, you will succeed only in condensing H₂O from the air, and risk ice contamination.

The total volume in your vial is about 1.0 mL at this point.

If you obtain a solution immediately, pipette the entire sample into your NMR tube using the same Pasteur pipette. Cap the NMR tube and invert it several times to mix.

If you don't have a solution, cap the vial and swirl it, but don't let the solution touch the cap, which is a possible source of contamination. Even a clean new cap can cause problems since many liners are glued to the cap and the glue can dissolve. You can use the Pasteur pipette to hasten solution by drawing up and releasing the suspension. Don't let the solution touch the rubber bulb. If you still don't have a solution after one minute, pipette the supernatant into the NMR tube. If you do this, be careful to avoid 2 phases in the NMR tube. Two liquid phases gives an NMR consisting of both phases superimposed because the tube spins during acquisition fast enough (at 20 Hz) to pull the top phase down into the bottom phase region via a vortex. For a solid compound, a solid floating about in a solution reduces resolution.

Do not use adhesive labels of any kind. If the adhesive gets to the inside of the probe, it will cause samples to not spin. Instead, push your tube through a piece of paper with the sample ID, or use tags with string. NMR tubes are reusable, but you might as well throw the caps away. Store your clean tubes horizontally in a glassware drying oven on aluminum foil, not in a beaker. In a beaker they will slowly become eccentric and you need them to be as round as possible.

Do not attempt to prepare a sample by adding the deuterated solvent to your compound in an NMR tube. Aside from contaminating the NMR solvent pipette, if you fail to obtain a solution, it will be difficult to shake the mixture out of the small diameter tube for phase separation.

Carry the NMR tube to the NMR room in an Erlenmeyer flask. If you attempt to carry the tube alone in your hand, you can easily drop it since it has no rough surfaces for gripping. A dropped NMR tube is a broken NMR tube; they don't survive.

Summary:

1. Clean dry vial.
2. Clean dry cap for vial.
3. Clean dry Pasteur pipette and spatula.
4. Clean dry rubber bulb.
5. Add 0.8 mL chloroform-d/TMS to your empty vial.
6. Add 10 drops distilled liquid, or 50 mg solid, to vial containing solvent.
7. Immediate soln: add to NMR tube with same Pasteur pipette. Cap and invert several times to mix.
8. No immediate soln: cap and swirl in vial. Do not let soln touch cap.
9. Push successful NMR tube through paper label or use string tag label. No adhesive label.
10. Use for ¹H and ¹³C spectra.