Appendix C

NMR SOLUTIONS AND TUBES

1. NMR tubes

The thin-walled glass NMR tubes that we use are 7 inches long with outside diameter of 5 millimeters. Relatively inexpensive generic tubes are suitable only for use in the probe of the Anasazi EFT 60 MHz FT-NMR. These tubes should never be used in the Varian Inova 400 MHz FT-NMR because they could cause damage to the probe. A more expensive tube, e. g., the Wilmad 507-PP or equivalent or better, is used in the 400 MHz instrument. These meet the necessary camber (convexity) specifications for the probe. The tubes are fragile. They most often break during cleaning when tops are snapped off by the spout of a wash bottle. Some plastic caps fit very tightly and so tubes may be broken if caps are not removed carefully. Occasionally, tubes are broken by being snapped off while inserting or removing them from the spinner. Tubes with a break near the top may be cut off and used, so these damaged tubes should not be discarded.

2. NMR solvents

The most commonly used NMR solvent is deuterated chloroform, $CDCl_3$. This and other solvents often are purchased with the reference compound for 0 ppm, tetramethyl silane (TMS), already added. The $CDCl_3$ used in our laboratories generally contains 0.05% TMS and about 0.2% $CHCl_3$. Thus, a small signal for the proton of $CHCl_3$ is frequently observed at about chemical shift 7.28 ppm. The carbon in $CDCl_3$ gives a triplet (often quite large in dilute samples) at about 77-78 ppm in the ^{13}C spectrum. This signal is a triplet due to the coupling between ^{2}D and ^{13}C nuclei. Note that in the case of deuterium, the n + 1 rule is more accurately expressed as the 2nI + 1 rule where I is nuclear spin. When the nucleus with which coupling occurs is ^{1}H, I = 1/2. However, when the nucleus is ^{2}D, I = 1, so 2nI +1 = 3 verifying that ^{13}C in $CDCl_3$ is a triplet. Available NMR solvents also include deuterated dimethyl sulfoxide, $(CD_3)_2SO$; deuterium oxide, D_2O; and many other deuterated solvents. Often a compound for analysis with insufficient solubility in $CDCl_3$ alone is brought into solution with the addition of a small amount of $(CD_3)_2SO$ to the $CDCl_3$.

3. Solution preparation

The volume of solution for NMR analysis must be sufficient to fill the tube to a depth of 6 cm or more for analysis with the 400 MHz instrument. This volume is about 650 µL. Variable depths may require frequent shimming to avoid poor 400 MHz spectra. Because of the excellent sensitivity of the Fourier transform instruments, very little sample compound is needed to obtain high quality spectra. As little as 1 or 2 mg may be satisfactorily analyzed. However, fewer transients and therefore shorter analysis times are required if solutions are somewhat more concentrated. In general we use 3-5 drops of a liquid sample and 10-20 mg of a solid sample with 650 µL of solvent. Volumes may measured with automatic pipets for good reproducibility and to conserve expensive solvents and sometimes limited amounts of sample. Generally solutions can be prepared directly in the NMR tubes. Thorough mixing of a newly prepared solution is essential. This is accomplished by inverting the capped tubes several times.

4. Cleanup

Containers for NMR solutions that are no longer needed are available in the hoods. Tubes are cleaned by rinsing first with acetone and then with diethyl ether. Rinsed tubes are inverted in a rack to drain. Before rinsed tubes are used again, they are pumped-out in an unheated vacuum oven. This ensures that no traces of acetone or diethyl ether remain in the tubes. Never use hot air to dry the tubes. Never use water, hot or cold, in the tubes.