The high-field spectrometers and the NMR service

These notes introduce the high-field NMR facilities in the Chemistry Research Laboratory and explain how to use the high-field and multinuclear service. The service complements the open-access facilities offered by the 200 & 400 MHz spectrometers.

1) NMR Facilities in the Chemistry Research Laboratory

The CRL currently houses a total of fifteen Fourier transform NMR spectrometers:

Organic Chemistry/Chemical Biology:

200 MHz:

Bruker DPX200 with manual sample insertion for open-access routine proton spectra

400 MHz:

Bruker AVIII400 ("AVF400") with sample changer (open-access ^{1}H, ^{13}C, DEPT, ^{19}F, ^{31}P, COSY and HSQC).
Bruker AVIII400 ("AVG400") with sample changer (open-access ^{1}H, ^{13}C, DEPT, ^{19}F, ^{31}P, COSY and HSQC).
Bruker AVIII400 ("AVH400") with sample changer (open-access ^{1}H, ^{13}C, DEPT, ^{19}F, ^{31}P, COSY and HSQC).
Bruker AVIII400 ("AVB400") with sample changer and broadband probe (trained users).

500 MHz: These run 1D and 2D homonuclear and heteronuclear experiments including more advanced studies

Bruker AVIII500 ("AVB500") spectrometer with ^{1}H/broadband and ^{1}H/^{19}F/^{13}C probes, sample changer and VT.
Bruker AVIII500 ("AVX500") spectrometer with broadband and triple resonance probes, sample changer and VT.

600 MHz:

Bruker AVIII600 ("AV600") spectrometer with a broadband Cryoprobe & sample changer.
Bruker NEO600 spectrometer with cryoprobe, sample changer and VT.

700 MHz:

Bruker AVIII700 spectrometer with a ^{1}H/^{13}C/^{15}N Cryoprobe & sample changer.

Inorganic Chemistry:

400 MHz:

Bruker AVIII400 spectrometer ("Hg400"), with broadband probe and sample changer
Bruker AVIII400 spectrometer ("Venus400"), with broadband probe, sample changer and VT.

500 MHz:

Bruker AVIII500 spectrometer ("AVD500"), with broadband probe and sample changer

Solid-state NMR 400 MHz:

Bruker HXY AVIII400 Widebore
Bruker HFX AVIII400 Widebore

These facilities are supported by five members of staff:

Organic/Chemical Biology:

Dr Harry Mackenzie: Head of Organic and Biological NMR. Office 00.120, Tel 75620/75659
Dr Coral Mycroft: NMR Service Manager. Office 00.120, Tel 75658/75659
Edward Tomlinson: NMR Research Technician. Office 00.123, Tel 75661/75659
Caitlin Salter: NMR Research Technician. Office 00.123, Tel 75661/75659 *(on leave)*

Inorganic:

Dr Nick Rees: Head of the Inorganic & Solid-State NMR facilities. Office 00.097A, Tel 85064/75659

2) The CRL NMR Submission Service

The NMR Submission service is available for all research workers in Organic Chemistry/Chemical Biology and makes use of the 500, 600 and 700 MHz instruments. Samples may be submitted for analysis to the service via the online booking system, leaving completed forms in the folder on the server, and depositing samples via the hatch into the basement High-Field NMR lab (00.086). Samples are later collected from the same place. You may seek the assistance of the NMR staff for the analysis of your data as necessary. This service is *always* very busy, so prior to submitting your sample for analysis a few considerations are worthwhile. Are the experiments you are asking to be run *really* necessary, and are they going to give you the information you require? If in doubt, ask a member of the NMR staff. Is the sample you are submitting of a quality suitable for high-field analysis (see under sample preparation)?

To submit a sample to the NMR service, you must fill out a submission form, which is self-explanatory, and enter details on the web-page. **All experiments required must be listed on the submission form itself, not just on the**

web-page. You **must** submit the 200/400 MHz spectrum/spectra of your sample with the submission form, to show that the sample is suitable for analysis. ***Part IIs must have their forms approved and signed by their laboratory supervisors before submission***. It is also necessary to indicate on the form if you expect to observe signals well outside the "usual" shift regions, that is 0-10 ppm for proton (e.g. acids peaks, metal hydrides) and 0-240 ppm for carbon (e.g. silyl derivatives, alkyl iodides). Completed data can be found on the NMR data archive on the **files-support** server.

All samples for the NMR service must be supplied in precision 5 mm tubes (at least a Wilmad 507-PP or equivalent model) which are **not** cracked, broken or otherwise damaged in any way, and ***must*** be at least 7" long.

3) Hands-on use of the high-field spectrometers

Research workers may also ask to be trained to use the high-field facilities themselves. Generally, this is available to D.Phil. and Post-doctoral workers (but also some Part IIs) who should initially seek approval from their supervisor and then contact the NMR Staff.

***Initial training must be given by a member of the departmental NMR staff, not by members of your, or another, research group*.**

You will be asked to undergo a training session regardless of whether you have used NMR instrumentation elsewhere. To book instrument time, use the online booking system for each spectrometer. The NEO600 is reserved for NMR staff use. Each instrument has certain periods reserved for use by the NMR service and research. The AV700 and AV600 are reserved mainly for specific biological projects.

4) Sample Preparation for NMR

These notes are intended to help you get the best out of the laboratory's NMR facilities by ensuring that the samples you analyse, or submit to the NMR service, are suitable for the experiments you require. The NMR service operates at full capacity, so it is of utmost importance that time is not wasted running samples which give unsatisfactory spectra due to careless preparation. A little time and effort in sample preparation will be repaid by consistently high quality spectra.

Sample tubes

High-field NMR requires the use of sample tubes of accurate dimensions, so called *precision tubes*. Higher quality models (Wilmad 507 or greater) must be used. These are available from the stores. **Tubes with broken tops must not be submitted to the NMR service**; they are potentially dangerous and may not be handled by the robot sample changers. It is important to keep tubes free from dust, grease etc., but they should *never* be cleaned with chromic acid, as these leave behind paramagnetic impurities in the glass which makes them unusable. The commercial cleaning agent "Decon-90" is a very effective alternative for removing contamination, especially grease. After soaking in a suitable solution, tubes can be rinsed with distilled water and then acetone. It is, however, surprisingly difficult to free 5 mm tubes from traces of acetone. An effective method is to blow nitrogen or air through the tube, with a pipette, while warming it gently for a few minutes or leave under vacuum. ***Do not*** subject NMR tubes to high temperatures for long periods of time (this includes oven drying) as this will cause them to distort and become unusable, and always ***lay tubes flat*** in the oven when drying.

Solvents

All NMR spectrometers use a deuterium lock system for field stabilisation, so it is essential to use solvents containing deuterium. The only common solvent excluded by the requirement is carbon tetrachloride. All the usual organic solvents are available in deuterated form, although some are rather expensive. It is wise to consider whether a cheaper alternative will suffice (chloroform, DMSO and D_2O are the cheapest solvents). Stocks of solvents are kept in the stores and are available for general use. These solvents are of medium quality with respect to levels of deuteration; for proton work with small quantities of sample, higher grade solvents sealed in 0.5 ml vials can be purchased from suppliers. When working with proton spectra of dilute samples, e.g. sub-milligram quantities, always consider where the signal from residual protonated solvent will occur. For solvents other than chloroform, the signal will be a multiplet due to coupling with deuterium and may fall in a rather inconvenient place. Another "impurity" signal to be aware of is that of water, which occurs in all solvents and often gives rise to a rather broad signal. Tables of some useful properties of common solvents are given at the back of this handout whilst tables of ^{1}H and ^{13}C shifts of common solvent impurities can be found in *J. Org. Chem*., 1997, **62**, 7512-7515 and *Organometallics*, 2010, **29**, 2176-2179 (these are extremely useful references: every lab should have a copy!)

Sample preparation

When preparing solutions for NMR spectroscopy, care should be taken to ensure cleanliness of apparatus and absence of contamination by solvent impurities. If you are surprised by giant peaks around 0.5-1.5 ppm (hydrocarbons) or around 0.1 ppm (silicone grease, which always seems to work its way off ground glass joints and into NMR samples), then reconsider your experimental procedures. When you have dissolved your sample, it may be advisable to filter the solution through a plug of cotton wool straight into the NMR tube to remove any particulate matter. Lack of solid particles is an **essential** requirement for obtaining high-quality spectra. Use the correct volume of solvent, as indicated on the sample submissions forms. Too much solvent dilutes the sample, reduces sensitivity and so wastes instrument time whereas too little solvent makes locking and shimming difficult or impossible.

Samples that are of insufficient quality will be returned for clean-up and must be resubmitted to the service. Clearly, this will lead to delays in obtaining your spectrum, so it is in your best interest to submit a sample worthy of high-field analysis.

5) Some cautionary notes

This section documents a number of points that can lead to wastage of instrument and operator time. Please bear these in mind when undertaking or requesting NMR analysis.

a) ***Sample quality***: Prior to submitting a sample for high-field analysis, **always** check the quality of the sample by running a 200/400 MHz spectrum first. Even if you have a weak sample, this spectrum will indicate the presence of solvent impurities or other unexpected contaminants. This spectrum should also be submitted with the sample when you request high-field analysis, to indicate that the quality of the submitted specimen is sufficient to warrant further studies.

b) ***Sample strength***: For high-field proton analysis, samples of up to *ca*. 10 mgs in 0.5 ml are sufficient - anything significantly more will increase solution viscosity and degrade resolution. For carbon spectra, sensitivity is the key factor and as much sample as is available should be used.

c) ***Heteronuclear Analysis***: Only submit very weak samples for high-field heteronuclear analysis e.g. ^{13}C, ^{31}P, etc when the sample has been shown to be of suitable quality and correct structure (where possible) by e.g. proton NMR. Instrument time is wasted by people who request long acquisitions for weak samples which turn out to be incomprehensible mixtures with meaningless spectra.

d) ***DEPT spectra***: Request these only if the multiplicity-edited HSQC experiment cannot provide you with the information you need (usually because of severe resonance crowding). HSQC provides more data than DEPT since it correlates attached protons, is very much faster to run (often minutes for HSQC versus hours for DEPT) and includes multiplicity editing equivalent to that found in DEPT.

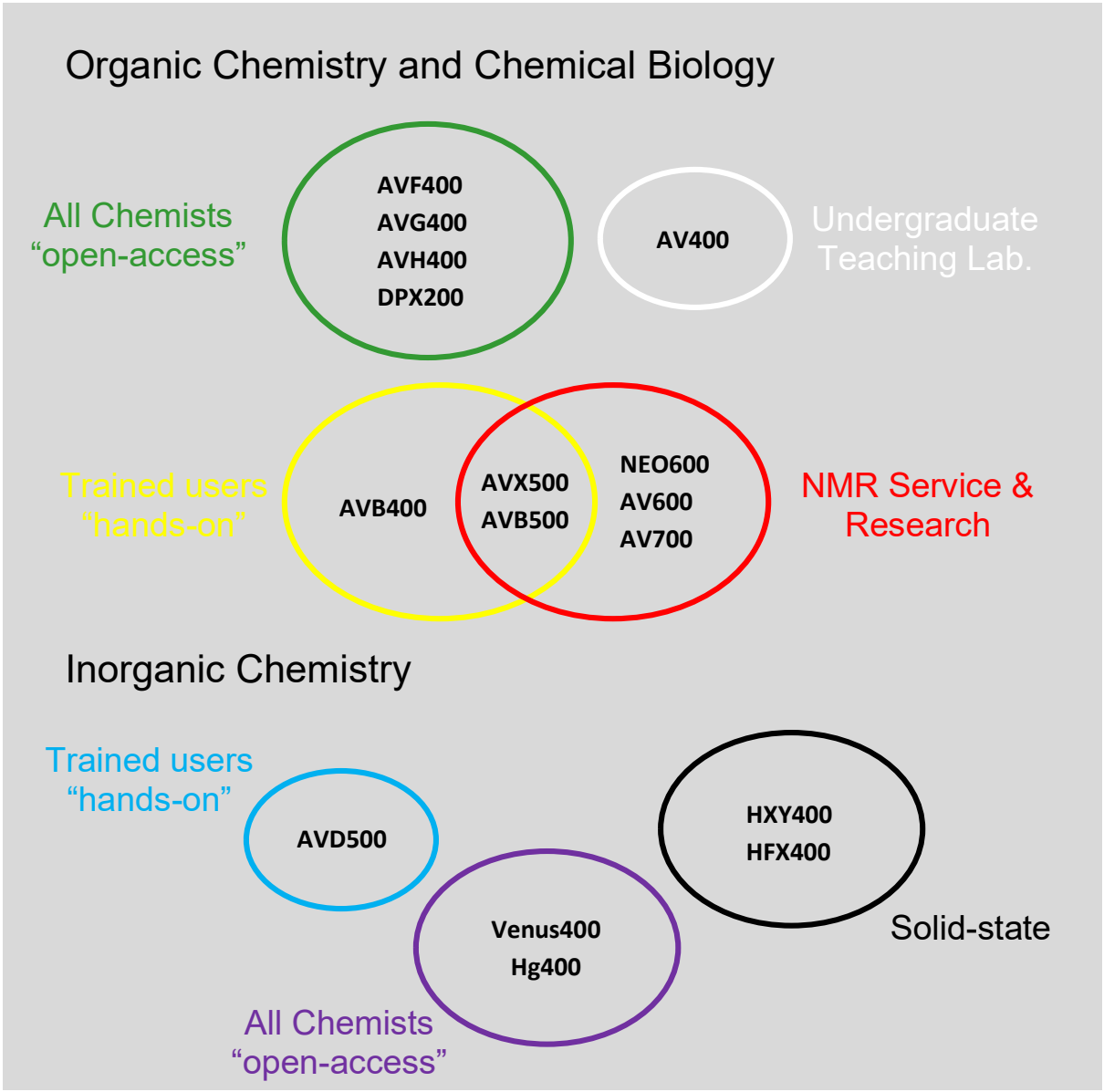
e) ***What do you really need?*:** Please refrain from asking for an extensive range of experiments to be run on one sample if it is not a final product or otherwise very important compound. Most information required can be derived from a careful choice of the necessary experiments. If you are in any doubt as to what these are for your particular problem, ask a member of the NMR staff, who should be able to advise you. Similarly, if you require specific data from some experiments e.g. accurate integration of proton spectra to determine product ratios, make a note of this on the submission form so that the data can be acquired and processed accordingly. The standard procedure followed for routine samples will not necessarily be optimum for the data you require. This is particularly relevant if you wish to interpret integration data accurately.

Properties of the commonly used deuterated NMR solvents

Solvent	δ_H /ppm	$\delta_{(HOD)}$ /ppm	δ_C /ppm	Melting Point/°C	Boiling Point/°C
Acetone-d_6	2.05	2.0	206.7, 29.9	-94	57
Acetonitrile-d_3	1.94	2.1	118.7, 1.4	-45	82
Benzene-d_6	7.16	0.4	128.4	5	80
Chloroform-d_1	7.26	1.5	77.0	-64	62
Deuterium oxide-d_2	4.80	4.8	-	3.8	101
Dichloromethane-d_2	5.32	1.5	54.0	-95	40
N,N-dimethyl formamide-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
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

NB. Proton shifts correspond to the residual partially protonated solvent.

Overview of Department of Chemistry NMR Facilities:



NMR Facility web pages QR code:

nmrweb.chem.ox.ac.uk