



# • TopSpin 3.x

Introduction to NMR Methods User Manual

Version 001

Innovation with Integrity

NMR Spectroscopy

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This manual was written by

Peter Ziegler

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Billerica, Massachusetts, USA

P/N: B7169

For further technical assistance on the TopSpin 3.x unit, please do not hesitate to contact your nearest BRUKER dealer or contact us directly at:

BRUKER Biospin Corporation 15 Fortune Drive Billerica, MA 01821 USA

Phone: (978) 667-9580 ext. 5444 FAX: (978) 667-2955 E-mail: applab@bruker-biospin.com Internet: www.bruker.com

1	Introdu	uction	9
	1.1	General	9
	1.2	Disclaimer	9
_	-		
2	Spectr	ometer Basics	11
	2.1	Introduction	11
	2.2	Magnetic Safety	11
	2.2.1	Safety Precautions within the Inner Zone	
	2.2.2	Safety precautions within the outer zone	
	2.3	Cryogenic Safety	13
	2.4	Electrical Safety	13
	2.5	Chemical Safety	
	2.6	CE Certification	
	2.7	AVANCE Architecture Overview	
	2.8	Sample preparation	15
	2.9	Inserting the Sample Plus Spinner into the Magnet	15
	2.10	Spinning the Sample	
	2.11	Tuning and Matching the Probe	17
	2.11.1	Probes equipped with ATM	
	2.11.1.1	Automatic tuning	17
	2.11.1.2	Manual tuning	
	2.11.2	Probes without ATM	
	2.12	Locking the sample	21
	2.13	Shimming the sample	21
	2.13.1	Shimming on the Lock Signal	
	2.13.2	Shimming on the FID (Free Induction Decay)	
	2.13.3	Shimming using the Tune file	
	2.13.4	Shimming using TopShim	
	2.14	Optimizing Resolution and Lineshape	
3	1-D Pro	oton experiment	25
	3.1	Sample	
	3.2	1-D Proton experiment	
	3.2.1	Introduction	
	3.2.2	Experiment setup	
	3.2.3	Acquisition	
	3.2.4	Processing	
	3.2.5	Optimizing the Spectral width	
	3.2.6	Plotting the 1D Proton spectra	
	3.2.7	Observations	
4	2-미 비스	monuclear experiments	27
-			

4.1	Sample	. 37
4.2	2-D gradient COSY	37
4.2.1	Introduction	.37
4.2.2	Preparation experiment	38
4.2.3	Setting up the COSY experiment	39
4.2.4	Acquisition	.41
4.2.5	Processing	42
4.2.6	Plotting	43
4.2.7	Observations	45
4.3	2-D gradient NOESY experiment	46
4.3.1	Introduction	.46
4.3.2	Preparation experiment	47
4.3.3	Setting up the NOESY experiment	47
4.3.4	Acquisition	50
4.3.5	Processing	50
4.3.6	Plotting	51
4.3.7	Observations	53
4.4	2-D phase sensitive TOCSY experiment	54
4.4.1	Introduction	54
4.4.2	Preparation experiment	55
4.4.3	Setting up the TOCSY experiment	55
4.4.4	Acquisition	. 58
4.4.5	Processing	.58
4.4.6	Plotting	59
4.4.7	Observations	61
1-D Se	lective experiments	63
5.1	Sample	63
5.2	1-D Selective COSY	.63
5.2.1	Introduction	.63
5.2.2	Reference spectrum	.64
5.2.3	Selective excitation region set up	65
5.2.3.1	On resonance	.65
5.2.4	Setting up the Selective COSY	66
5.2.5	Acquisition	68
5.2.6	Processing	68
5.2.7	Plotting two spectra on to the same page	71
5.2.8	Observations	72
5.3	1-D Selective NOESY	73
5.3.1	Introduction	.73
5.3.2	Reference spectrum	.73
5.3.3	Selective excitation region set up	74
5.3.3.1	On resonance	.74

5

## Table of Contents

5.3.4	Setting up the Selective NOESY	76
5.3.4	Setting up the Selective NOESY	7

	5.3.5	Acquisition	
	5.3.6	Processing	78
	5.3.7	Plotting two spectra on to the same page	80
	5.3.8	Observations	82
	5.4	1-D Selective TOCSY	83
	5.4.1	Introduction	83
	5.4.2	Reference spectrum	83
	5.4.3	Selective excitation region set up	84
	5.4.3.1	On resonance	84
	5.4.4	Setting up the Selective TOCSY	85
	5.4.5	Acquisition	87
	5.4.6	Processing	87
	5.4.7	Plotting two spectra on to the same page	90
	5.4.8	Observations	91
e	1 D C	arban avnarimenta	02
0	6 1		<b>93</b> 03
	6.2	1-D Carbon Experiment	
	6.2.1	Introduction	
	622	Experiment set up	94
	623	Acquisition	
	624	Processing	
	6.2.5	Plotting the 1D Carbon spectrum	
	6.2.6	Observations	
	6.3	DEPT-135 Experiment	
	6.3.1		
	6.3.2	Experiment set up	
	6.3.3	Acquisition	
	6.3.4	Processing	
	6.3.5	Observations	106
	6.4	DEPT-90 Experiment	107
	6.4.1	Introduction	107
	6.4.2	Experiment set up	107
	6.4.3	Acquisition	108
	6.4.4	Processing	109
	6.4.5	Observations	110
7	2_⊓ ⊔,	ateronuclear experiments	111
•	<b>2-D</b> HE		1 1 1
	7.1		 111
	7.2 7.2 1	Introduction	 111
	7.2.1	Proparation experiment	۱۱۱ 110
	1.2.2	Preparation experiment	1

	7.2.3	Setting up the HSQC experiment	113
	7.2.4	Acquisition	117
	7.2.5	Processing	117
	7.2.6	Plotting	
	7.2.7	Observations	120
	7.3	2D HMBC experiment	121
	7.3.1	Introduction	121
	7.3.2	Preparation experiment	121
	7.3.3	Setting up the HMBC experiment	
	7.3.4	Acquisition	126
	7.3.5	Processing	126
	7.3.6	Plotting	
	7.3.7	Observations	131
0	Deter	mination of 00 degree nulses	400
0		Introduction	133 133
	8.2	Proton 90 degree transmitter pulse	133
	821	Parameter setun	133
	822	Acquisition	136
	823	Processing	136
	824	Determine the 900 pulse	140
	825	Observations	144
	8.3	Carbon 90 degree transmitter pulse	
	8.3.1	Parameter setup	
	8.3.2	Acquisition	
	8.3.3	Processing	
	8.3.4	Determine the 900 pulse	
	8.3.5	Observations	
_			
9	Sensi	itivity tests	157
	9.1	Introduction	
	9.2	1H Sensitivity test	
	9.2.1	Experiment setup	
	9.2.2	Acquisition	
	9.2.3	Processing	
	9.2.4	Calculating the Signal to Noise ratio	
	9.2.5	Observations	
	9.3	13C Sensitivity test with 1H decoupling	
	9.3.1	Experiment setup	
	9.3.2	Acquisition	
	9.3.3	Processing	
	9.3.4	Calculating the Signal to Noise ratio	

## **Table of Contents**

	9.3.5	Observations
	9.4	13C Sensitivity test without 1H decoupling 172
	9.4.1	Experiment setup 172
	9.4.2	Acquisition
	9.4.3	Processing175
	9.4.4	Calculating the Signal to Noise ratio175
	9.4.5	Observations
10	Spect	rometer configuration
	10.1	Hardware Configuration 181
	10.1	Expinstall 186
	10.2	Set up the crop job for NMR save
	10.0	Selection of current Probehead 193
	10.4 1	Current probe equipped with pics: 193
	10.4.1	Current probe not equipped with pics and with probe parameters: 194
	10.4.2	Current probe not equipped with pics and with probe parameters:
	10.4.0	Lock File setup
	10.5 1	Setting the BSMS field 197
	10.5.1	Setting the Field compensation 199
	10.0.2	Observations 202
	10.0	
11	Hardw	are203
	11.1	Power up procedure for an AV-III console

	11.1	Power up procedure for an AV-III console	
	11.2	Resetting the ELCB board in the BSMS on a AV-II console	
	11.3	Downloading a new DRU Firmware	207
	11.4	Observations	
Α	Арре	endix	213
	A.1	Standard Parameter set list	213
	A.2	Standard Test Samples	
в	Cont	act	217

## **1** Introduction

## 1.1 General

This manual was written for AVANCE systems running TopSpin and should be used as a guide through the set up process for some experiments. The success of running the experiments in this manual is under the assumption that all parameters have been entered in to the prosol table.

## 1.2 Disclaimer

This guide should only be used for its intended purpose as described in this manual. Use of the manual for any purpose other than that for which it is intended is taken only at the users own risk and invalidates any and all manufacturer warranties.

Some parameter values, especially power levels suggested in this manual may not be suitable for all systems (e.g. Cryo probes) and could cause damage to the unit. Therefore only persons trained in the operation of the AVANCE systems should operate the unit.

#### B7169\_00\_01

## **2** Spectrometer Basics

## 2.1 Introduction

In terms of safety the presence of a relatively strong magnet is what differentiatesNMR spectrometers from most other laboratory equipment. When designing an NMR laboratory, or training personnel who will work in or around the laboratory, no other feature is of greater significance. As long as correct procedures are adhered to, working in the vicinity of superconductive magnets is completely safe and has no known harmful medical side effects. Negligence however can result in serious accidents. It is important that people working in the vicinity of the magnet fully understand the potential hazards. Of critical importance is that people fitted with cardiac pacemakers or metallic implants should never be allowed near the magnet.

The magnet is potentially hazardous due to:

- 1. The large attractive force it exerts on ferromagnetic objects.
- 2. The large content of liquid Nitrogen and Helium.

## 2.2 Magnetic Safety

A Magnetic Field surrounds the magnet in all directions. This field (known as the stray field) is invisible, hence the need to post warning signs at appropriate locations. Objects made of ferromagnetic materials, e.g. iron, steel etc. will be attracted to the magnet. If a ferromagnetic object is brought too close, it may suddenly be drawn into the magnet with surprising force. This may damage the magnet, or cause personal injury to anybody in the way!

Because the strength of the stray field drops significantly as one moves away from the magnet, it is useful to discuss safety in terms of two broadly defined regions, the inner and outer zone. In terms of organizing a laboratory as well as defining good work practices, the concept of an inner and outer zone is particularly useful.

The physical extent of these two zones will depend on the size of the magnet. The bigger the magnet, the stronger the stray magnetic fields and hence the larger the extent of the two zones. Figure 2.1. shows the concept of the two zones (not drawn to scale). Details of stray fields for various magnets can be found in the Site Planning Guides delivered with the BASH CD.





### 2.2.1 Safety Precautions within the Inner Zone

The inner zone extends from the magnet center to the 1mT (10 Gauss) line. Within this region objects may suddenly be drawn towards the magnet center. The attractive force of the magnet can change from barely noticeable to uncontrollable within a very short distance. **Under no circumstances should heavy ferromagnetic objects be located** 

#### or moved within this zone.

Any ladders used when working on the magnet should be made of non-magnetic material such as aluminum. Helium and nitrogen dewars which are used to top up the liquid levels inside the magnet must be made of non-magnetic material.

Do not allow small steel objects (screwdrivers, bolts etc.) to lie on the floor near the magnet. These could cause serious damage if drawn into the magnet bore, especially when no probe is inserted in the magnet.

Mechanical watches may be damaged if worn within the inner zone. Digital watches can be worn safely. Of course, the precautions for the outer zone which will now be discussed must also be adhered to within the inner zone.

#### 2.2.2 Safety precautions within the outer zone

The outer zone extends from the 1mT line to the 0.3mT line. The magnet's stray field does not get blocked by walls, floors or ceilings and the outer zone may well encompass adjoining rooms. The stray field may erase information stored on magnetic tapes or discs. Bank cards, security passes or any devices containing a magnetic strip may be damaged. CD's will not be damaged, although CD drives may contain magnetic parts. When using pressurized gas cylinders made of steel, they should be located well beyond the outer zone (preferably outside the magnet room) and must always be properly fixed to the wall. The color display of computer monitors may suffer some distortion when located too close to the magnet, although permanent damage is unlikely. Once beyond the outer zone any special precautions on account of the magnet stray field are no longer necessary.

## 2.3 Cryogenic Safety

The magnet contains relatively large quantities of liquid helium and nitrogen. These liquids, referred to as cryogens, serve to keep the magnet core at a very low temperature.

Because of the very low temperatures involved, **gloves**, a long sleeved shirt or lab **coat** and **safety goggles** should always be worn when handling cryogens. Direct contact with these liquids can cause frostbite. The system manager should regularly check and make sure that evaporating gases are free to escape from the magnet, i.e. the release valves must not be blocked. Do not attempt to refill the magnet with helium or nitrogen unless you have been trained in the correct procedure.

Helium and nitrogen are non-toxic gases. However, because of a possible **magnet quench**, whereupon the room may suddenly fill with evaporated gases, adequate ventilation must always be provided.

## 2.4 Electrical Safety

The spectrometer hardware is no more or less hazardous than any typical electronic or pneumatic hardware and should be treated accordingly. Do not remove any of the protective panels from the various units. They are fitted to protect you and should be opened by qualified service personnel only. The main panel at the rear of the console is designed to be removed using two quick release screws, but again, this should only be

done by trained personnel. Please note that, unless disconnected, cooling fans on the rear panel will continue to run even with the panel removed.

## 2.5 Chemical Safety

Users should be fully aware of any hazards associated with the samples they are working with. Organic compounds may be highly flammable, corrosive, carcinogenic etc.

## 2.6 **CE Certification**

All major hardware units housed in the AVANCE with SGU consoles as well as peripheral units such as the HPPR, shim systems, probe and BSMS keyboards comply with the CE Declaration of Conformity. This includes the level of any stray electromagnetic radiation that might be emitted as well as standard electrical hazards.

NOTE: To minimize electromagnetic radiation leakage, the doors of the console should be closed and the rear paneling mounted.

## 2.7 AVANCE Architecture Overview



Figure 2.2

NOTE: Please use the BASH (Bruker Advanced Service Handbook) for further information about the AVANCE system and hardware.

## 2.8 Sample preparation

- Use clean and dry sample tubes
- Use medium to high quality sample tubes
- Always filter the sample solution
- Always use the same sample volume or solution height
- Filling volume of a 5 mm tubes is 0.6 ml or 5 cm
- Filling volume of a 10 mm tubes is 4 ml or 5 cm
- Use the sample depth gauge to adjust the sample depth (1.8 cm for older style probes, 2.0 cm for newer style probes)
- The sample tube should sit tightly inside the spinner
- Wipe the sample tube clean before inserting into magnet
- Turn on lift air to insert the sample into the magnet

## 2.9 Inserting the Sample Plus Spinner into the Magnet

The raising and lowering of the sample is controlled by a stream of pressurized air. Be careful never to lift the sample with the plug still inserted at the top of the magnet bore. Newer BOSS-2 shim systems are designed not to enable the LIFT if the magnet bore is still plugged. Furthermore, make sure that the air flow is present (it is quite audible) before placing a sample onto the top of the bore.



To insert the sample plus spinner into the magnet use the following procedure:

1. If present, remove the plug from the top of the magnet bore

2. Activate the LIFT button on the BSMS keyboard. A flow of air will be heard and if a sample is already in the magnet it will be raised and suspended on a cushion of air at the top of the magnet bore.

3. Remove the old sample and place the new sample onto the air cushion

4. Press the LIFT key again. The sample will gently drop into the magnet and will settle at a precise position within the probe.

## 2.10 Spinning the Sample

A second function of pressurized air is to enable the sample to rotate. The spinning of the sample serves to "even-out" some of the inhomogeneities that may exist in the magnetic field at the center of the magnet.

NOTE: Sample tubes with a diameter of less then 5mm and samples to be investigated using inverse probes are normally not rotated.

#### Set the spin rate using the following procedure:

- 1. Open the BSMS display
- 2. Click on the SPIN button to activate the spinning.

#### Suggested spin rates are:

20 Hz for a 5 mm probe

12 Hz for a 10 mm probe

## 2.11 Tuning and Matching the Probe

The sensitivity of any probe will vary with the frequency of the signal transmitted to it and there exists a frequency at which the probe is most sensitive. Furthermore this frequency may be adjusted over a certain range using tuning capacitors built into the probe circuitry. **Tuning** involves adjusting the probe circuitry so that the frequency at which it is most sensitive is the relevant transmission frequency (SFO1, SFO2 etc.) Each coil in the probe will be tuned (and matched) separately.

If the probe has been changed or the transmission frequency altered significantly, it may be necessary to retune the probe. For routine work in organic solvents with selective probes, the value of the transmitted frequencies are unlikely to vary greatly. Hence, once the probe has been initially tuned, slight variations in frequency will not warrant retuning. Typically the transmitted frequency would need to be altered by at least 100kHz to warrant retuning. However for broadband probes the frequencies transmitted will vary greatly from nucleus to nucleus and so the probe will need to be tuned each time the selected nucleus is altered.

Whenever a probe is tuned it should also be matched. **Matching** involves ensuring that the maximum amount of the power arriving at the probe base is transmitted up to the coil which lies towards the top of the probe. This ensures that the minimum amount of the power arriving at the probe base is reflected back towards the amplifiers (and consequently wasted).

NOTE: Bruker offers two different types of Tuning and Matching adjustments. In addition to the manual adjustments of the tuning and matching capacitors, the probes can be equipped with a Automatic Tuning Module (ATM). Follow the steps below for either option.

#### 2.11.1 Probes equipped with ATM

#### 2.11.1.1 Automatic tuning

- 1. Type edc and create a new data set
- 2. Type atma

NOTE: The display will switch automatically to the acquisition window and displays the wobble curve. The tuning and matching is performed automatically. If multiple frequencies are used in a parameter set such as C13CPD, HNCACOGP3D etc., ATMA will start adjusting the lowest frequency first and will switch in the order of increasing frequency automatically.

#### 2.11.1.2 Manual tuning

#### 1. Type atmm

NOTE: The ATM control window appears (see Figure 2.4) and the display will switch automatically to the acquisition window and displays the wobble curve. (Figure 3.2).

#### Figure 2.4



3. Click on the **'Tuning'** buttons in the ATM control window to move the wobble curve in to the center of the display

4. Click on the '**Matching**' buttons in the ATM control window to adjust the dip of the wobble curve to the lowest position

NOTE: Since the Tuning and Matching adjustment interact with each other, a repeat of steps 3 and 4 are necessary for a perfect tune and match (see Figure 2.5). If multiple frequencies are used in a parameter set such as C13CPD, use the '**Nucleus Selection**' radio buttons in the ATM control window to switch to another nucleus and repeat steps 3 and 4.



#### 2.11.2 Probes without ATM

- 1. Type edc and create a new data set
- 2. Type wobb

NOTE: The display will switch automatically to the acquisition window and displays the wobble curve (see Figure 2.6). If multiple frequencies are used in a parameter set such as C13CPD, HNCACOGP3D etc., wobb will start with the lowest frequency first. The nuclei are selected in the order of increasing frequency. Tuning and Matching rods are color-coded for different nuclei e.g. yellow for 1H, blue for 13C etc.

3. Adjust the Tuning rod marked **T** underneath the probe to move the wobble curve in to the center of the display

4. Adjust the Matching rod marked  ${\bf M}$  underneath the probe to adjust the dip of the wobble curve to the lowest position

NOTE: Since the Tuning and Matching adjustment interact with each other, a repeat of steps 3 and 4 are necessary for a perfect tune and match (see Figure 2.6). If multiple frequencies are used in a parameter set and to switch to other frequencies, follow steps 5 through 6 below. and repeat steps 3 and 4.





- 5. Click on 📶 in the acquisition window
- Figure 2.7







6. Click 💼 on to terminate the acquisition

## 2.12 Locking the sample

To display the lock signal type **lockdisp** on the Topspin command line. This opens a window in which the lock trace appears.

The most convenient way to lock is to use the TopSpin command lock. To start the lockin procedure, enter lock and select the appropriate solvent from the menu. Alternatively, enter the solvent name together with the lock command, e.g., lock cdcl3. During lock-in, several parameters such as the lock power, the field value, and the frequency shift for the solvent are set according to the values in the lock table. This table can be edited using the command edlock. Note that the lock power listed in this table is the level used after the sample has been locked. The field-shift mode is then selected and autolock is activated. Once lock-in is achieved, the lock gain is set so that the lock signal is visible in the lock window. At this point the message "lock: finished" appears in the status line at the bottom of the window.

The lock-in procedure outlined above sets the frequency shift to the exact frequency shift value for the given solvent as listed in the edlock table. It also sets the field value to the value listed in the edlock table and then adjusts it slightly to a given ppm value no longer depends on the lock solvent). Following this lock-in procedure, the solvent parameter in the eda table is set automatically, which is important if you wish to use the automatic calibration command sref (see "Spectrum Calibration and Optimization").

The lock-phase adjustment by monitoring the sweep wiggles (i.e., while the field is not locked but is being swept) is recommended each time the probehead is changed, because autolock may fail. If the original phase is reasonably close to the correct value, lock-in can be achieved and the phase can be adjusted using autophase. Note that the lock phase for each probehead is stored in the edlock table. In some cases, the lock power level listed in the edlock table is set too high leading to a saturation of the lock signal. Usually, lock-in can be achieved, but the signal oscillates due to saturation. A quick fix is simply to reduce the lock power manually after lockin. However, it is better to change the power level in the edlock table.

NOTE: that the appropriate lock power level depends on the lock solvent, the field value, and the probehead. Any value changes in the edlock table should only be done by experts.

## 2.13 Shimming the sample

The following is intended to be a practical guide for adjusting the room temperature shim system (BOSS). The purpose of shimming is to maximize the magnetic field homogeneity, which depends somewhat on probehead and sample geometry. In general, it is necessary to shim the magnetic field after each probehead change, sample change, and occasionally between changes to correct for any system drifts.

Optimal shim settings may vary substantially from probehead to probehead; however, provided the probehead is always positioned the same in the magnet and the sample is always positioned the same with respect to the receiver coil, the shim values for a given probehead will be fairly reproducible. Thus, shimming time can be greatly reduced if the shim settings for each probehead are stored as a shim file on the computer. When the probehead is changed, the shim file for the new probehead can be read in and then final adjustments can be made to these shim values to correct for system drifts, and to

account for the geometry of the particular sample being used.

The BOSS shim system consists of a number of shim coils arranged in the room temperature bore of the magnet. During shimming, the currents in these shim coils are adjusted so that the small magnetic field gradients produced cancel the residual inhomogeneity of the main magnetic field (H0) as completely as possible.

#### 2.13.1 Shimming on the Lock Signal

When the spectrometer is locked, the vertical offset of the lock trace on the graphics display corresponds to the amplitude of the lock substance signal, assuming constant lock DC, gain, and power levels. The lock level, then, serves as useful guide for basic shim adjustment. The goal in shimming on the lock signal is to adjust the shims so that the lock trace appears as high on the graphics display as possible. This lock level corresponds to the highest possible lock substance signal amplitude.

#### 2.13.2 Shimming on the FID (Free Induction Decay)

The shape of the FID, and especially the beginning of the FID, indicates the shape of the transformed signal line, while the length of the FID tail is important to the overall resolution. For good line shape and high resolution, the shim controls must be adjusted so that the FID envelope is truly exponential with the longest possible decay time.

#### 2.13.3 Shimming using the Tune file

This method of shimming is useful when gradients are not available. A simple text file is edited to give the BSMS the instructions to shim the sample automatically. A default shim file "example\_bsms" can be edited using the edtune command and then stored with a new name in <TopSpin-home>/exp/stan/nmr/lists/group. The file and can be executed with the command tune. Figure 2.9 shows an example of a tune file.

Figure 2.9

# SHIMMIT spi	in	
MAXLOCK	0.4	
TIMEOUT	1800	
LOCKDWELL	3	
# Shim_name	Maximum_Step_Size	Number_of_Iterations
Z	30	3
Z2	30	3
Z	5	5
Z2	5	5
Z3	5	5

### 2.13.4 Shimming using TopShim

TopShim is a tool designed for easy and automatic shimming. It requires that the instrument is equipped with a gradient amplifier and a gradient probe. In addition a 2HTX board or any 2H amplifier is necessary for Deuterium gradient shimming.

The core method of TopShim is gradient shimming. This is complemented by a spectrum optimization approach, where a quality criterion for the final lineshape ensures the best results for all possible situations.

Both 1D and 3D shimming modes are provided to adjust only the on-axis or both the onand off-axis shim functions, respectively. However 3D is restricted to only Proton gradient shimming using a H2O sample.

The acquisition of the B0 field map data can be carried out with 1H or 2H observation, enabling the use of TopShim for protonated as well as deuterated solvents.

Optionally the additional tune functionality can be applied before and/or after gradient shimming in order to adjust low order shims for maximum lock level.

For further information please consult the TopSpin Automatic Shimming Users Manual in the Help section of TopSpin.

## 2.14 Optimizing Resolution and Lineshape

The standard sample for measuring the proton lineshape and resolution specifications is, CHCL3 in Acetone-d6. The concentration of CHCL3 depends on the field strength of the magnet and the probe and can vary from 3% down to 0.1%.

For measuring the 13C resolution and lineshape test the standard sample ASTM (60% Dioxane in 40% C6D6) sample may be used.

For both tests the line shape is measured at 50%, 055% and 0.11% of the peak. The Bruker standard parameter sets to use for this tests are PRORESOL and C13RESOL.

Figure 2.10 and 2.11 are illustrating the influence of the On-axis shims on the lineshape.

Figure 2.10



#### Figure 2.11



Adjusted Spectrum

## **3 1-D Proton experiment**

## 3.1 Sample

A sample of **30mg Menthyl Anthranilate in DMSO-d6** is used for the experiment in this chapter

Figure 3.1



## 3.2 1-D Proton experiment

#### 3.2.1 Introduction

Section 3.2 describes the acquisition and processing of a one-dimensional 1H NMR spectrum using the standard Bruker parameter set **PROTON**. The pulse sequence **zg30**, Figure 3.2 consists of the recycling delay, the radio-frequency (RF) pulse, and the acquisition time during which the signal is recorded. The pulse angle is shown to be 30 degrees. The two parameters, D1 and P1, correspond to the length of the recycle delay, and the length of the 90 degree RF pulse, respectively.

Figure 3.2



The time intervals depicted in the pulse sequence diagrams are not drawn to scale. For

example, d1 is typically a few seconds while p1 is typically a few microseconds in length.

#### 3.2.2 Experiment setup

1. Click on the ' <b>Sta</b>	rt' tab in the	TopSpin M	enu bar			
Figure 3.3						
<u> </u>	uire <u>P</u> rocess	A <u>n</u> alyse	P <u>u</u> blish	<u>V</u> iew	Manage	0
	Create Dataset	📕 Find Datase	t 🕥 Ope	n <u>D</u> ataset	Paste	Dataset 🖹 Read Pars.
2. Select Crea	ate Dataset	by clicking	g on it			
3. Enter the followi	ng informatio	n in to the	'New' w	ndow		
Figure 3.4						
	New					X
	Initializing its NMR For multi-receiver Please define the NAME EXPNO PROCNO DIR Solvent Experiment Dirs. Experiment TITLE	Parameters acc experiments sev number of recein proton_exp 1 1 C:\data3.0 C:\Bruker\Top	ording to the veral dataset vers in the b Spin3.0.b.40 PROTC	DMS N	xperiment typ ed. :O mr\par	xe. ▼ ▼ ▼ ▼ ▼ ▼ ▼ ▼
	1D- Proton expe 30 mg Menthyl / Show new da	eriment Anthranilate in Di ataset in new wind (1,2,16) OK	MSO-d6 dow Cancel	More Info	o Help	

NOTE: The directory (DIR) is specific to how the data are stored and therefore may show different entries as the one in Figure 3.4 above. Click on the down arrow button to browse for a specific directory.

4. Click on OK

5. Click on the 'Aquire' tab in the TopSpin menu bar

Figure 3.5 Start Acquire Process Analyse Publish View Manage Sample 
Gov Options 
Gov Options 
Gov Options

6. Select 🙀 Sample 🗸 by clicking on it

Figure	3.6
--------	-----



#### 7. Select 'ej' by clicking on it

NOTE: Wait till the sample lift air is turned on and remove any sample which may have been in the magnet.

8. Place the sample on too the top of the magnet

9. Select 💐 Sample 🔻 by clicking	on i	it
----------------------------------	------	----

Figure 3.7



#### 10. Select 'ij' by clicking on it

NOTE: Wait till the sample is lowered down in to the probe and the lift air is turned off. A licking sound may be heard.



Figure 3.8

△ Solvent	Description	1
Acetic	acetic acid-d4	
Acetone	acetone-d6	
Acetone_Hump	acetone-d6 (CHCl3)	
C6D6	benzene-d6	
CD2CI2	methylenechloride-d2	
CD3CN	acetonitrile-d3	
CDCI3	chloroform-d	
CDCI3_SENS	chloroform-d (ETB)	
CH3CN+D2O	HPLC Solvent (Acetonitril/D2O)	
D20	deuteriumoxide	
DEE	diethylether-d10	
Dioxane	dioxane-d8	
DME	dimethylether-d6	
DMF	dimethylformamide-d7	
DMSO	dimethylsulfoxide-d6	
EtOD	ethanol-d6	
H2O+D2O	90%H2O and 10%D2O	1

12. Select 'DMSO' by clicking on it

13. Select V Tune 🗸 by clicking on it

NOTE: This performs a 'atma' (automatic tuning) and requires a probe equipped with a automatic tuning module. Other options can be selected by clicking on the down arrow inside the 'Tune' button.

15. Select	clicking on it	
Figure 3.9		
		Turn sample rotation on (ro on)
		Turn sample rotation off (ro off)
		Change sample rotation rate (ro)
		MAS Pneumatic Unit (masdisp)

16. Select 'ro on' by clicking on it

NOTE: Rotation may be turned off for probes such as BBI, TXI, TBI and for small sample probes.

17. Select	Shim ▼	by clicking on it
NOTE: This arrow insid	s executes the e the ' <b>Shim</b> ' b	e command ' <b>topshim</b> '.To select other options. click on the down utton.
18. Select	<mark>∬</mark> Pr <u>o</u> sol	by clicking on it

NOTE: This will load the pulse width and power levels in to the parameter set.

#### 3.2.3 Acquisition



#### 3.2.4 Processing

1. Click on the 'Process' tab in the TopSpin Menu bar

#### Figure 3.10

•											
		<u>S</u> tai	rt <u>A</u> co	luire	Process	A <u>n</u> alys	e P <u>u</u> blish	<u>V</u> iew	<u>M</u> anage	2	
		A Pro	<u>c</u> . Spectr	um 🔻	Adjust F	Phase 🗢	Å Calib. A <u>x</u> is -	✓ <sup>*</sup> P	ick P <u>e</u> aks <del>√</del>	∫ <u>I</u> ntegrate <del>→</del>	A <u>d</u> vanced <del>▼</del>
2. (	Click	c on 🛛	ΛPr	'0 <u>c</u> .	Spectru	m 🔻					

NOTE: This executes a processing program including commands such as an exponential window function 'em', Fourier transformation 'ft', an automatic phase correction 'apk' and a baseline correction 'abs'. Other options are available by clicking on the down arrow inside the 'Proc. Spectrum' button.



### 3.2.5 Optimizing the Spectral width

- 1. Click on
- 3. Type the following F1 [ppm] values:

From = 9

To = -1





8. Expand the spectrum to include all peaks



NOTE: This enters the manual Integration mode. Other options are available by clicking on the down arrow inside the '**Integrate**' button.

10. Set the cursor line, starting at the left of the spectrum, to the left of the first peak to be integrated, click the left mouse button and drag the cursor line to the right of the peak, then release the mouse button





11. Repeat step13 for the remainder of the peaks



12. Click on **[**] to save the integration regions

## 3.2.6 Plotting the 1D Proton spectra

- 1. Expand the spectrum (all peaks in display)
- 2. Click on
- 3. Type the following F1 [ppm] values:

From = **8.5** 

To = **0.5** 

Figure 3.18

💐 exactzoom	
Please enter of the desired	the exact coordinates d expansion.
	F1 [ppm]
From	8.5000
То	0.5000
	OK Cancel

## 4. Click on OK

5. Click on the 'Publish' tab in the TopSpin Menu bar

```
Figure 3.19
```

	<u>S</u> tart	<u>A</u> cquire	Process	A <u>n</u> alyse	P <u>u</u> blish	<u>V</u> iew	<u>M</u> anage	0
			<u> </u>	py 🗳 P <u>r</u> int		t Layout <del>v</del>		E-Mail

6. Click on **□ Plot Layout** <del>▼</del>



NOTE: If desired, any changes can be administered by clicking on the icon to open the Plot Editor.

7. Click on the to plot the spectrum

## 3.2.7 Observations

#### B7169\_00\_01
# 4 2-D Homonuclear experiments

# 4.1 Sample

A sample of **30mg Menthyl Anthranilate in DMSO-d6** is used for the experiments in this chapter

Figure 4.1



# 4.2 2-D gradient COSY

#### 4.2.1 Introduction

The COSY experiment relies on the J-coupling to provide spin-spin correlation, and whose cross peaks indicate which 1H's are close to which other 1H's through the bonds of the molecule. Typically proton which are up 3 bonds away can be observed.

The signals acquired with one of these experiments have absorptive and dispersive line shape contributions in both F1 and F2 dimensions. This means that it is impossible to phase the spectrum with all peaks purely absorptive, and, as a consequence, the spectrum must be displayed in magnitude mode. A typical spectral resolution of 3 Hz/pt is sufficient for resolving large scalar couplings. In order to resolve small J-couplings fine digital resolution is required, which significantly increases the experimental time. In general, the DQF-COSY experiment is recommended if a higher resolution is desired.

Using pulsed field gradients (PFG), the coherence pathway selection and the axial peak suppression can be achieved with only one scan per time increment. Thus, if enough substance is available, a typical gradient COSY experiment with 128 time increments can be recorded in 5 minutes.

Section 4.2 describes the acquisition and processing of a two-dimensional 1H gradient COSY. The standard Bruker parameter set is **COSYGPSW** and includes the pulse sequence **cosygpppqf** shown in Figure 4.2. It consists of the recycling delay, two radio-

frequency (RF) pulses, separated by the increment delay D0 and the acquisition time during which the signal is recorded. Both pulses have a 90 degrees angle. Two gradient pulses are applied before and after the second pulse in the sequence. Purge pulses are applied before d1.





The time intervals depicted in the pulse sequence diagrams are not drawn to scale. For example, d1 is typically a few seconds while p1 is typically a few microseconds in length.

#### 4.2.2 **Preparation experiment**

1. Run a **1D Proton** spectrum, following the instructions in **Chapter 3, 1-D Proton experiment, Paragraph 3.2.2 Experiment setup** through **3.2.4 Processing**.





## 4.2.3 Setting up the COSY experiment

1. Click on the 'Star	rt' tab in the T	ГорSpin M	lenu bar					
Figure 4.4								
<u>S</u> tart <u>A</u> cqu	ire <u>P</u> rocess	A <u>n</u> alyse	P <u>u</u> blish	<u>V</u> iew	<u>M</u> anage	2		
	reate Dataset	Find Datase	et 🔄 Ope	n <u>D</u> ataset	🚺 Paste I	Dataset	Read	Pars.
2. Select Crea	te Dataset	by clicking	g on it					
3. Enter the followir	ng information	n in to the	'New' wi	ndow				
Figure 4.5								
riguio 4.0	New					3		
	Prepare for a new initializing its NMR For multi-receiver Please define the NAME	experiment by parameters acc experiments se number of rece proton_exp	creating a ne cording to the everal dataset ivers in the b	w data set a selected e: s are create ox below.	and xperiment type ed.			
	EXPNO	2				_		
	DIR	1 C:\data3	0					
	Solvent	0.100100.	•	DMS	0	-		
	Experiment Dirs.	C:\Bruker\Top	Spin3.0.b.40	\exp\stan\nr	mr\par	-		
	Experiment TITLE		COSYG	PSW	•	~		
	2D- gradient CO 30 mg Menthyl A Show new dat	SY experiment inthranilate in D taset in new win (1,2,16)	0MSO-d6 ndow					
		ОК	Cancel	More Info	Help			

NOTE: The directory (DIR) is specific to how the data are stored and therefore may show different entries as the one in Figure 4.5 above. Click on the down arrow button to browse for a specific directory.

4. Click on OK

5. Click on the 'Aquire' tab in the TopSpin menu bar



Turn sample rotation on (ro on)
Turn sample rotation off (ro off)
Change sample rotation rate (ro)
MAS Pneumatic Unit (masdisp)

7. Select 'ro off' by clicking on it

NOTE: 2-D experin	nents should be run non spinning
8. Select APro	sol by clicking on it
NOTE: This will loa	d the pulse width and power levels in to the parameter set.
9. Select 🔲 SetL	imits by clicking on it
Figure 4.8	
	setlimits 🛛
	<ul> <li>Close this dialog box after setting frequencies.</li> <li>1. Open 1D dataset from Browser.</li> <li>2. Zoom into region of interest.</li> <li>3. Click OK to set frequencies and return to original dataset.</li> </ul>

10. To open the 1D Proton spectrum, right click on the dataset name in the browser window (e.g. proton\_exp 1) and select 'Display' or click and hold the left mouse button for dragging the 1D Proton dataset in to the spectrum window

OK

Cancel

11. Expand the spectrum to display all peaks, leaving ca. 0.2 ppm of baseline on either side of the spectrum

NOTE: The solvent peak may be excluded if it falls outside of the region of interest. Digital filtering however is only applied in F2 and the solvent peak is folding in F1.



NOTE: The display changes back to the 2D data set.

# 4.2.4 Acquisition



#### 4.2.5 Processing



NOTE: This executes a standard processing program **proc2**. The message shown in Figure 4.12 pops up in case of a magnitude 2D experiment and the apk2d option is enabled. To configure the processing program follow the steps below.

3. Click on the down arrow	inside the <mark> </mark>	button
Figure 4.13		_
	Configure Standard Processing (proc2d)	
	Process F2+F1 Axis (xfb)	
	Process Only F2 Axis (xf2)	
	Process Only F1 Axis (xf1)	
	Symmetrize Spectrum (sym)	
	Start Automation AU Program (xaup)	
		1

3. Select 'Configure Standard Processing' by clicking on it



NOTE: To avoid the message shown in Figure 4.12 the option 'Auto-Phasing (apk2d)' may be disabled for magnitude like 2D experiment.



#### 4.2.6 Plotting

- 1. Use the the buttons to adjust for a suitable contour level
- 2. Click on the 'Publish' tab in the TopSpin Menu bar

#### Figure 4.16

	<u>S</u> tart	<u>A</u> cquire	<u>P</u> rocess	A <u>n</u> alyse	P <u>u</u> blish	<u>V</u> iew	<u>M</u> anage	0
			<u> </u>	py 🗳 P <u>r</u> in	t 🗢 🖂 P <u>l</u> o	t Layout <del>~</del>	<u> </u>	E-Mail

6. Click on Brand Plot Layout

7. Select the 'Plot' tab by clicking on it

Figure 4.17

rum ProcPars Acqui	Pars Title	PulseProg	Peaks	Integrals	Sample	Structure	Plot	Fid Ac	qu
🎾 Auto 🏡 🎴 +/20	)_hom.xwp								
2D- gradient COS	V experime	nt.						-	-
30 mg Menthyl An	thranilate	in DMS0-da	5					BPÍ	
						- h		()	$\sim$
							ppm	Current Data	Farameters proton_eap
li l					1			FI - Angulai	1 ILLON FAILURETEERS
					a	að °		TLDI TLDI DESTROPI PROMAD 5 1	20100230 14.38 sp051 up.DUL 13C-1
			(H		<b>P</b> \$3	₩.	1	FULFENCE TO SOLVENT RS	2049 2049 0400
4			89		1 6	酃。		os Sala Viceaci AO	0 2400,748 Hg 1,172250 Hg 0,4203001 Hg
1			6		87	₩ P4	2	pro CN CN TE	64 208.247 UARD 4.30 UARD 283.1 H
-					0			00 01 011	0.00000300 ant 2.0000000 ant 0.03000000 ant
				Å			3	013 014 180	0,0000400 ass 0.00020000 ass 0.00041455 ass
				Y				HUCI FQ	In 10.50 used
				0			4	F1 FLNI FLNI FLNI	10.10 uses 2500.00 uses 22.4139333 N 3.69430000 N
			8	a	8 g	8,		GPEARS	SHEQIC.ICO
						17 - 199 <b>.</b> 2	5	PII PII FI - Acquis	10,00 % 1000,00 uses 11100 persectors
								TD SPOL FICHES SN	128 200,1312 HHz 18,754002 Hz 7,999 ppp
							6	PRIOCE F2 - Process S1	QF Ling parameters 1024
- The second sec	• • •							17 509 518 0 18 0 1	100.1100000 8842 Q0186
-		1		٥			7	GL 0 FC FL - Fronse	L.40
-	606	1				, a		11 90.2 17	1014 GP 300.1300000 MHz
							in the second	218 0 18 0 i	-

NOTE: If desired, any changes can be administered by clicking on the icon to open the Plot Editor.

8. Click on the **end** to plot the spectrum

# 4.2.7 Observations

# 4.3 2-D gradient NOESY experiment

#### 4.3.1 Introduction

NOESY (Nuclear Overhauser Effect SpectroscopY) is a 2D spectroscopy method used to identify spins undergoing cross-relaxation and to measure the cross-relaxation rates. Most commonly, NOESY is used as a homonuclear 1H technique. In NOESY, direct dipolar couplings provide the primary means of cross-relaxation, and so spins undergoing cross-relaxation are those which are close to one another in space. Thus, the cross peaks of a NOESY spectrum indicate which protons are close to each other in space. This can be distinguished from COSY, for example, which relies on J-coupling to provide spin-spin correlation, and whose cross peaks indicate which 1H's are close to which other 1H's through the bonds of the molecule.

The basic NOESY sequence consists of three p/2 pulses. The first pulse creates transverse spin magnetization. This precesses during the evolution time t1, which is incremented during the course of the 2D experiment. The second pulse produces longitudinal magnetization equal to the transverse magnetization component orthogonal to the pulse direction. Thus, the basic idea is to produce an initial situation for the mixing period d8. Note that, for he basic NOESY experiment, d8 is kept constant throughout the 2D experiment. The third pulse creates transverse magnetization from the remaining longitudinal magnetization. Acquisition begins immediately following the third pulse, and the transverse magnetization is observed as a function of the time t2. The NOESY spectrum is generated by a 2D Fourier transform with respect to t1 and t2.

Axial peaks, which originate from magnetization that has relaxed during tm, can be removed by the appropriate phase cycling.

NOESY spectra can be obtained in 2D absorption mode. Occasionally, COSY-type artifacts appear in the NOESY spectrum; however, these are easy to identify by their antiphase multiplet structure.

Section 4.3 describes the acquisition and processing of a two-dimensional 1H phase sensitive NOESY. The standard Bruker parameter set is **NOESYPHSW** and includes the pulse sequence **noesygpphpp** shown in Figure 4.18. It consists of the recycling delay, three radio-frequency (RF) pulses, separated by the increment delay D0 between the first and second pulse, a mixing time D8 between the second and third pulse and the acquisition time during which the signal is recorded. All three pulses are of 90 degree.



The time intervals depicted in the pulse sequence diagrams are not drawn to scale. For example, d1 is typically a few seconds while p1 is typically a few microseconds in length.

# 4.3.2 Preparation experiment

1. Run a **1D Proton** spectrum, following the instructions in **Chapter 3, 1-D Proton experiment, Paragraph 3.2.2 Experiment setup** through **3.2.4 Processing**.



## 4.3.3 Setting up the NOESY experiment

1. Click on the 'Start' tab in the TopSpin Menu bar



Figure 4.21

NAME	proton_exp			
EXPNO	2			
PROCNO	1			
DIR	C:\data3.0	0		1
Solvent			DMSO	~
Experiment Dirs.	C:\Bruker\Top	Spin3.0.b.40\e	p\stan\nmr\par	•
Experiment TITLE		COSYGPS	W	1
2D- gradient CO 30 mg Menthyl A	SY experiment Inthranilate in DI taset in new wind	MSO-d6 low		

NOTE: The directory (DIR) is specific to how the data are stored and therefore may show different entries as the one in Figure 4.5 above. Click on the down arrow button to browse for a specific directory.

- 4. Click on OK
- 5. Click on the 'Aquire' tab in the TopSpin menu bar



NOTE: 2-D experiments should be run non spinning

8. Select AProsol by clicking on it



10. To open the 1D Proton spectrum, right click on the dataset name in the browser window (e.g. proton\_exp 1) and select 'Display' or click and hold the left mouse button for dragging the 1D Proton dataset in to the spectrum window

11. Expand the spectrum to display all peaks, leaving ca. 0.2 ppm of baseline on either side of the spectrum

NOTE: The solvent peak may be excluded if it falls outside of the region of interest. Digital filtering however is only applied in F2 and the solvent peak is folding in F1.



12. Click on OK to a	ssign the new limit	
Figure 4.26		
	H spectral limits copied for F1 and F2 dimensions. SW: 7.9997 ppm O1P: 4.024 ppm	
	Close	
13. Click on Close		
NOTE: The display chang	ges back to the 2D data set.	
14. Select the ' <b>AcquPars</b>	' tab by clicking on it av the pulse program parameters	
16. Make the following ch	ange	
Figure 4.27		
D8 [sec]	0.450	Mixing time
NOTE: The mixing time	depends on the size of the Molecule. The re	ange for Biomele

NOTE: The mixing time depends on the size of the Molecule. The range for Biomolecules are typically from 0.05 to 0.2 sec., medium size molelecules from 0.1 to 0.5 s ec. and for small molecules 0.5 to 0.9 sec.

17. Select the 'Spectrum' tab by clicking on it

## 4.3.4 Acquisition



## 4.3.5 Processing

1. Click on the 'Process' tab in the TopSpin Menu bar

	<u>S</u> tart	<u>A</u> cquire	Process	A <u>n</u> alys	e P <u>u</u> blish	<u>V</u> iew	<u>M</u> anage	0	
	∫	spectrum <del>v</del>	Adjust F	'hase 🗢	Å Calib. A <u>x</u> is च	• 👯 F	Pick P <u>e</u> aks <del>⇒</del>	∫ <u>I</u> ntegrate <del>~</del>	A <u>d</u> vanced <del>▼</del>



NOTE: This executes a standard processing program **proc2**. To configure this program or select other options, click on the down arrow inside the '**Proc. Spectrum**' button.



# 4.3.6 Plotting

- 1. Use the the buttons to adjust for a suitable contour level
- 2. Click on the '**Publish**' tab in the TopSpin Menu bar



- 6. Click on Brief Plot Layout
- 7. Select the 'Plot' tab by clicking on it



NOTE: If desired, any changes can be administered by clicking on the icon to open the Plot Editor.

8. Click on the <u>Man</u> to plot the spectrum

# 4.3.7 Observations

# 4.4 2-D phase sensitive TOCSY experiment

#### 4.4.1 Introduction

Figure 4.32

TOCSY (TOtal Correlation SpectroscopY) provides a different mechanism of coherence transfer than COSY for 2D correlation spectroscopy in liquids. In TOCSY, cross peaks are generated between all members of a coupled spin network. An advantage is that pure absorption mode spectra with positive intensity peaks are created. In traditional COSY, cross peaks have zero integrated intensity and the coherence transfer is restricted to directly spincoupled nuclei. In TOCSY, oscillatory exchange is established which proceeds through the entire coupling network so that there can be net magnetization transfer from one spin to another even without direct coupling. The isotropic mixing which occurs during the spin-lock period of the TOCSY sequence exchanges all in-phase as well as antiphase coherence.

The coherence transfer period of the TOCSY sequence occurs during a multiple-pulse spin-lock period. The multiple-pulse spin-lock sequence most commonly used is MLEV-17. The length of the spin-lock period determines how far the spin coupling network will be probed. A general rule of thumb is that 1/(10 JHH) should be allowed for each transfer step, and five transfer steps are typically desired for the TOCSY spectrum.

Section 4.4 describes the acquisition and processing of a two-dimensional 1H phase sensitive TOCSY. The standard Bruker parameter set is **MLEVPHSW** and includes the pulse sequence **mlevphpp** shown in Figure 4.14. It consists of the recycling delay, two radio-frequency (RF) pulses, separated by the increment delay D0 and the acquisition time during which the signal is recorded. The first RF pulse is a 90 degree pulse, the second pulse is the mlev spinlock pulse.



The time intervals depicted in the pulse sequence diagrams are not drawn to scale. For example, d1 is typically a few seconds while p1 is typically a few microseconds in length.

# 4.4.2 Preparation experiment

1. Run a **1D Proton** spectrum, following the instructions in **Chapter 3, 1-D Proton** experiment, Paragraph 3.2.2 Experiment setup through 3.2.4 Processing.



## 4.4.3 Setting up the TOCSY experiment

1. Click on the 'Start' tab in the TopSpin Menu bar



Figure 4.35

NAME	proton_exp			
EXPNO	4			
PROCNO	1			
DIR	C:\data3.0	כ		-
Solvent	1		DMSO	-
Experiment Dirs.	C:\Bruker\Top	Spin3.0.b.40	exp\stan\nmr\par	*
Experiment		MLEVPH	ISW	*
TITLE				
2D- TOCSY expe 30 mg Menthyl A	eriment Inthranilate in D taset in new win	MSO-d6		

NOTE: The directory (DIR) is specific to how the data are stored and therefore may show different entries as the one in Figure 4.35 above. Click on the down arrow button to browse for a specific directory.

- 4. Click on OK
- 5. Click on the 'Aquire' tab in the TopSpin menu bar



NOTE: 2-D experiments should be run non spinning

8. Select AProsol by clicking on it



10. To open the 1D Proton spectrum, right click on the dataset name in the browser window (e.g. proton\_exp 1) and select 'Display' or click and hold the left mouse button for dragging the 1D Proton dataset in to the spectrum window

11. Expand the spectrum to display all peaks, leaving ca. 0.2 ppm of baseline on either side of the spectrum

NOTE: The solvent peak may be excluded if it falls outside of the region of interest. Digital filtering however is only applied in F2 and the solvent peak is folding in F1.



NOTE: This will load the pulse width and power levels in to the parameter set.

12. Click on OK	to assign the new limit	
Figure 4.40		_
	E 🛛	
	1H spectral limits copied for F1 and F2 dimensions.	
	U SW: 7.9997 ppm	
	O1P: 4.024 ppm	
	Close	
13. Click on Close		
NOTE: The display of	changes back to the 2D data set.	
14. Select the 'Acqu	Pars' tab by clicking on it	
15. Click on 👖 to	display the pulse program parameters	
16. Make the following	ng change	
Figure 4.41		
D9 [sec]	0.0800000	TOCSY mixing time
NOTE: A mixing time	∋ of 0.06 to 0.08 sec. is typically for the TOCSY	' experiment.
17 Select the ' <b>Snec</b>	trum' tab by clicking on it	
TT. Delect the <b>Opec</b>		
<b>A</b> i=i(i=		
Acquisition		
1. Select Gain	by clicking on it	
2. Select Bos	by clicking on it	
Processing		
1. Click on the 'Proc	ess' tab in the TopSpin Menu bar	
Figure 4.42		
<u> </u>	e / Process \ Analyse Publish View Manage ② ▼ Adjust Phase ▼ Adjust Phase ▼ Adjust Phase ▼ ∫ In	tegrate <del>▼</del> A <u>d</u> vanced <del>▼</del>
2 Salaat	by dicking on it	
2. Select Proc.	Spectrum v by clicking on it	

4.4.4

4.4.5

NOTE: This executes a standard processing program **proc2**. To configure this program or select other options, click on the down arrow inside the '**Proc. Spectrum**' button.



# 4.4.6 Plotting

- 1. Use the isolations to adjust for a suitable contour level
- 2. Click on the 'Publish' tab in the TopSpin Menu bar



- 6. Click on Plot Layout -
- 7. Select the 'Plot' tab by clicking on it



NOTE: If desired, any changes can be administered by clicking on the icon to open the Plot Editor.

8. Click on the <u>Man</u> to plot the spectrum

# 4.4.7 Observations

#### B7169\_00\_01

# **5 1-D Selective experiments**

# 5.1 Sample

A sample of **30mg Menthyl Anthranilate in DMSO-d6** is used for the experiment in this chapter

Figure 5.1



# 5.2 1-D Selective COSY

#### 5.2.1 Introduction

The hard pulses used in all the experiments from the previous chapters are used to uniformly excite the entire spectral width. This chapter introduces soft pulses which selectively excite only one multiplet of a 1H spectrum. Important characteristics of a soft pulse include the shape, the amplitude, and the length. The selectivity of a pulse is measured by its ability to excite a certain resonance (or group of resonances) without affecting near neighbors. Since the length of the selective pulse affects its selectivity, the length is selected based on the selectivity desired and then the pulse amplitude (i.e., power level) is adjusted to give a 90° (or 270°) flip angle.

NOTE: The transmitter offset frequency of the selective pulse must be set to the frequency of the desired resonance. This transmitter frequency does not have to be the same as o1p (the offset frequency of the hard pulses), but for reasons of simplicity, they are often chosen to be identical.

Most selective excitation experiments rely on phase cycling, and thus subtraction of spectra, to eliminate large unwanted signals. It is important to minimize possible sources of subtraction artifacts, and for this reason it is generally suggested to run selective

experiments using pulse field gradients and non-spinning.

Section 5.2 describes the acquisition and processing of a one-dimensional 1H selective gradient COSY experiment. The standard Bruker parameter set is **SELCOGP** and includes the pulse sequence **selcogp** shown in Figure 5.2. It consists of the recycling delay, four radio-frequency (RF) pulses and the acquisition time during which the signal is recorded. The first RF pulse is a 90 degree pulse, followed by a 180 degree shaped pulse, a 180 degree hard pulse and finally a 90 degree pulse. The delay between the 180 and 90 degree pulse is 1/4\*J(H,H). The gradient pulses are applied before and after the shape pulse.

Figure 5.2



#### 5.2.2 Reference spectrum

1. Run a **1D Proton** spectrum, following the instructions in **Chapter 3**, **1-D Proton** experiment, **3.2.2 Experiment setup**, through **3.2.4 Processing** 





#### Selective excitation region set up 5.2.3

#### 5.2.3.1 On resonance

NOTE: Make sure that the SW is large enough to cover the entire Spectrum accounting for the position of O1. The shaped pulse is applied on resonance (at the O1 position) The power level and width of the excitation pulse have to be known and entered into the Prosol parameter table

1. Type wrpa

Figure 5.4

Copy data set. will be a 1-file d Please specify	If NAME ends with ".top", the destin ataset (no expno/procno required). destination:
NAME =	sel_cosy
EXPNO =	1
PROCNO =	1
DID -	C:\data3.0

2. Change NAME = sel\_cosy

3. Click on OK		
4. Type re		
Figure 5.5		
	💩 re	
	Options Oisplay data in sat Display data in ne	me window w window
	NAME =	sel_cosy
	EXPNO =	1
	PROCNO =	1
	DIR =	C:\data3.0
	OK Cancel	Browse Find Help

- 5. Change NAME = sel\_cosy
- 6. Click on OK
- 7. Expand peak at 7.7 ppm
- 8. Click on to set the RF from cursor



9. Move the cursor line in to the center of the multiplet

10. Click the left mouse button to set the frequency

Figure 5.7

🍓 01/02/03	
Define SFO1/O1 f	requencies
SFO1 [MHz] =	300.132307
O1/2/3 [Hz] =	2307.41
01 02	O3 Cancel

11. Click on 01

# 5.2.4 Setting up the Selective COSY

1. Click on the 'Start' tab in the TopSpin Menu bar

F	igure 5.8	8									
		<u>S</u> tart	<u>A</u> cquire	Process	A <u>n</u> alyse	P <u>u</u> blish	<u>V</u> iew	<u>M</u> anage	2		
			C <u>r</u> eate	Dataset	📕 F <u>i</u> nd Datas	et 🕥 Ope	en <u>D</u> ataset	Paste	Dataset	Read Pars.	
2	. Sele	ct 🔝	Read P	ars. by	clicking or	n it					

#### Figure 5.9

💐 Parameter Sets: rp	ar			X
File Options He	elp	Source = C:V	Bruker\TopSpin3.0.b.4	2\exp\stan\nmr\par
Find file names 🛛 🖌	enter any string, *, 1	?		
Class = Any	Dim = 🔽 🗆 S	Show Recommended		
Type = 🔄 SubTy	pe = Rese	et Filters		
SELCO1H	SELCOGP	SELGPSE	SELMLGP	SELMLZF1H
SELNO1H	SELNOGP	SELRO1H	SELROGP	SELU 13C
SELU_1H	SELU_COSY	SELU_HMBC	SELU_HSQC	SELU_ROESY
SELZG1H				
				Read Close

NOTE: Enter **SEL**\* in to the '**Find file names**' window and hit '**Enter**' to display all selective parameter sets shown in figure 5.11.

- 3. Select 'SELCOGP'
- 4. Click on Read...
- 5. Select the acqu, proc and outd parameter options only
- 6. Click on the down arrow next to the 'Keep the following parameter' window
- 7. Select 'P1, O1, PLW1' from the pull down menu

Figure 5.10

Ocurre Descurator Oct. ONDerland	
Source Parameter Set = C:\Bruker\	TopSpin3.0.b.42/exp\stan\nmr\par\SELCC
Destination Data Set = selective_ex	(p 2 1 C:\data3.0
<ol> <li>Select the desired file types of th</li> </ol>	ie source parameter set
<ol><li>Press OK to copy them to the de</li></ol>	stination data set.
acqu	
proc	
outd	
title	
Keen the following parameters:	
Keep the following parameters:	
Keep the following parameters: P 1, 01, PLW 1	
Keep the following parameters: P 1, 01, PLW 1	

- 8. Click on OK
- 9. Select the 'Title' tab by clicking on it

10. Make the following changes:

1-D Selective gradient COSY experiment 30 mg Menthyl Anthranilate in DMSO-d6

- 11. Click on 📳 to store the title
- 12. Select the 'Spectrum' tab by clicking on it

13. Click on the 'Aquire' tab in the TopSpin menu bar

Figure 5.11	
<u>Start</u> <u>A</u> cquire <u>P</u> roces	s A <u>n</u> alyse P <u>u</u> blish <u>V</u> iew <u>M</u> anage 🕜
🕴 Sample 🚽 🏧 Lock 🕅 T	une 🔻 🔱 Spin 👻 🛱 Shim 👻 🕂 Prosol 💹 Gain 👻 🕨 Go 👻 Options 👻
Select IL Spin by click	king on it
Figure 5.12	
	Tu <u>r</u> n sample rotation on (ro on)
	Turn sample rotation off (ro off)
	Change sample rotation rate (ro)
	MAS Pneumatic Unit (masdisp)

7. Select 'ro off' by clicking on it

NOTE: 1-D selective experiments should be run non spinning

8. Select **AProsol** by clicking on it

NOTE: This will load the pulse width and power levels in to the parameter set.

#### 5.2.5 Acquisition



#### 5.2.6 Processing

1. Click on the 'Process' tab in the TopSpin Menu bar

Fig	Figure 5.13								
	<u>Start</u> <u>A</u> cquire <u>P</u> rocess A <u>n</u> alyse	P <u>u</u> blish <u>V</u> iew <u>M</u> anage 🕜							
	Λ Pro <u>c</u> . Spectrum ▼ Adjust Phase ▼	🕅 Calib. A <u>x</u> is 👻 🎊 Pick P <u>e</u> aks 🗢 🗍 Integrate 🗢 Advanced 🗢							
2. Fig	2. Click on the down arrow inside the <b>A Proc. Spectrum</b> button								
	Configure S	andard Processing (proc1d)							
Window M <u>u</u> ltiplication (wm) Fourier <u>T</u> ransform (ft)									
	Sta <u>r</u> t Autom	ation AU Program (xaup)							

- 3. Select 'Configure Standard Processing' by clicking on it
- 4. Deselect the following options:

'Auto-Phasing (apk)' 'Set Spectrum Reference (sref)' 'Auto-Baseline correction (abs)' 'Warn if Processed data exist'

Figure 5.15

Press 'Execute' to process the cur Press 'Save' to just change the pro Changed options will be effective v one-click 'Proc. Spectrum' button.	rent ( ocess when	dataset. sing options. pressing the	
Exponential Multiply (em)	•	LB [Hz] =	0.1
Fourier Transform (ft)			
Auto - Phasing (apk)			
Set Spectrum Reference (sref)			
Auto - Baseline Correction (abs)			
Plot (autoplot)		LAYOUT =	+/1D_H.xwp
Marp if proceeded data swiet			

5. Click on Execute

6. Expand the spectrum from 8 ppm to 6 ppm



7. Click on 😽 Adjust Phase 🐱

8. Adjust the **0** order phase on the peak at 6.5 ppm to display a antiphase pattern



# 9. Click on 🔄 to store the phase value

# 5.2.7 Plotting two spectra on to the same page

- 1. Display the selective COSY spectrum
- 2. Click on kiele to enter the Multiple display option
- 3. Drag the Reference spectrum in to the spectral window





4. Click on the 'Publish' tab in the TopSpin Menu bar

Figure 5.19

8										
		<u>S</u> tart	<u>A</u> cquire	<u>P</u> rocess	A <u>n</u> alyse	P <u>u</u> blish	<u>V</u> iew	<u>M</u> anage	2	
				<u> </u>	opy 🗳 P <u>r</u> in	t 🗢 🕒 P <u>l</u> o	t Layout 🔻	PDF ▼	E-Mail	
5.	Click o	n the	Print v	button	to print the	e active w	indow			

# 5.2.8 Observations
# 5.3 1-D Selective NOESY

#### 5.3.1 Introduction

Section 5.4 describes the acquisition and processing of a one-dimensional 1H selective gradient NOESY experiment. The standard Bruker parameter set is **SELNOGP** and includes the pulse sequence **selnogp** shown in Figure 5.20. It consists of the recycling delay, five radio-frequency (RF) pulses and the acquisition time during which the signal is recorded. The first RF pulse is a 90 degree pulse, followed by a 180 degree shaped pulse, a 90 degree pulse, a 180 degree pulse and finally a 90 degree pulse. The mixing time **D8** is applied before and after the 180 degree pulse. There are four gradient pulses applied, one each between the RF pulses.

Figure 5.20



#### 5.3.2 Reference spectrum

1. Run a **1D Proton** spectrum, following the instructions in **Chapter 3**, **1-D Proton** experiment, **3.2.2 Experiment setup**, through **3.2.4 Processing** 



# 5.3.3 Selective excitation region set up

## 5.3.3.1 On resonance

NOTE: Make sure that the SW is large enough to cover the entire Spectrum accounting for the position of O1. The shaped pulse is applied on resonance (at the O1 position) The power level and width of the excitation pulse have to be known and entered into the Prosol parameter table

1. Type wrpa

Figure 5.22

Copy data set. I will be a 1-file da Please specify o	f NAME ends with ".top", the destinatior ataset (no expno/procno required). Jestination:	
NAME =	sel_noesy	
EXPNO =	1	
PROCNO =	1	
	Culdate 2.0	

2. Change NAME = sel\_noesy

3. Click on OK 4. Type re		
Figure 5.23	🙀 re	X
	Options O Display data in sat O Display data in ne	me window w window
	NAME = EXPNO = PROCNO = DIR =	sel_noesy 1 1 C:\data3.0
	OK Cancel	Browse Find Help

- 5. Change NAME = sel\_noesy
- 6. Click on OK
- 7. Expand peak at 4.8 ppm
- 8. Click on 🚺 to set the RF from cursor

#### Figure 5.24



- 9. Move the cursor line in to the center of the multiplet
- 10. Click the left mouse button to set the frequency

🤤 01/02/03
Define SFO1/O1 frequencies
SFO1 [MHz] = 300.131438
O1/2/3 [Hz] = 1437.87
O1 O2 O3 Cancel

# 5.3.4 Setting up the Selective NOESY

1. Click or	n the ' <b>Start</b> '	tab in the Top	Spin Menu bar			
Figure 5.26						
<u> </u>	tart <u>A</u> cquire	<u>P</u> rocess A <u>r</u>	alyse P <u>u</u> blish	<u>V</u> iew <u>M</u> anage	ə 🕜	
	C <u>r</u> ea	te Dataset 🛛 🔙 F <u>i</u> r	d Dataset 🔄 Ope	en <u>D</u> ataset 🚺 Pas	ste Dataset 🔣 Read Pars.	
2. Select	🔡 Read F	Pars. by click	king on it			
Figure 5.27						
	Parameter Sets: rp	ar				
Fil	le Options He	əlp	Source = C:\Bru	Iker\TopSpin3.0.b.42\	exp\stan\nmr\par 🖌	
Fin	Find file names venter any string, *, ?					
Clas	Class = Any Vim = Vin Show Recommended					
Тур	e = 🔽 SubTy	pe = 💉 Reset I	Filters			
SEL	LCO1H	SELCOGP	SELGPSE	SELMLGP	SELMLZF1H	
SEL	LNO1H	SELNOGP	SELRO1H	SELROGP	SELU_13C	
SEL	LU_1H 7C1H	SELU_COSY	SELU_HMBC	SELU_HSQC	SELU_ROESY	
JOL L	2011					
					Read Close	
Fil Fin Clas Typ SEL SEL SEL	Parameter Sets: rp le Options He d file names ess = Any ess ess = Any ess e = SubTyp conH LVO1H LU_1H LZG1H	ar ar arter any string, *, ? Dim = Dim = Shope = Reset i SELCOGP SELNOGP SELU_COSY	Source = C:\Bru	Iker\TopSpin3.0.b.42\ SELMLGP SELROGP SELU_HSQC	SELMLZF1H SELU_13C SELU_ROESY	

NOTE: Enter **SEL**\* in to the '**Find file names**' window and hit '**Enter**' to display all selective parameter sets shown in figure 5.29.

- 3. Select 'SELNOGP'
- 4. Click on Read...
- 5. Select the following parameter options: acqu, proc, outd

- 6. Click on the down arrow next to the 'Keep the following parameter' window
- 7. Select 'P1, O1, PLW1' from the pull down menu

Figure 5.28	
	🔄 rpar 🛛 🔀
	Source Parameter Set = C:\Bruker\TopSpin3.0.b.42\exp\stan\nmr\par\SELCOGP Destination Data Set = selective_exp 2 1 C:\data3.0 1) Select the desired file types of the source parameter set 2) Press OK to copy them to the destination data set.
	acqu
	proc outd
	title
	Keep the following parameters:
	P 1, 01, PLW 1
	OK Cancel Keep

8. Click on OK

9. Select the 'Title' tab by clicking on it

10. Make the following changes:

1-D Selective gradient NOESY experiment 30 mg Menthyl Anthranilate in DMSO-d6

- 11. Click on The to store the title
- 12. Select the 'AcquPars' tab by clicking on it
- 13. Click on the 'Aquire' tab in the TopSpin menu bar



7. Select 'ro off' by clicking on it

NOTE: 1-D selectiv	e experiments should be run non s	pinning		
8. Select <b>Prosol</b> by clicking on it				
NOTE: This will load the pulse width and power levels in to the parameter set.				
9. Select the ' <b>Acqu</b>	Pars' tab by clicking on it			
10. Make the follow	ing parameter changes:			
Figure 5.51				

# 5.3.5 Acquisition

1. Select Gain by clicking on it
NOTE: To adjust rg manually, click on the down arrow inside the 'Gain' icon
2. Select Select by clicking on it
NOTE: Other options are available by clicking on the down arrow inside the ' <b>Go</b> ' button.

# 5.3.6 Processing

1. Click on the 'Process' tab in the TopSpin Menu bar Figure 5.32 0 Start Acquire Process A<u>n</u>alyse Publish View Manage Λ Proc. Spectrum ▼ Adjust Phase ▼ Advanced ▼ 2. Click on the down arrow inside the Figure 5.33 Configure Standard Processing (proc1d) Window Multiplication (wm) Fourier Transform (ft) Start Automation AU Program (xaup)

3. Select 'Configure Standard Processing' by clicking on it

4. Deselect the following options:

'Auto-Phasing (apk)' 'Set Spectrum Reference (sref)' 'Auto-Baseline correction (abs)' 'Warn if Processed data exist'

Figure 5.34

Press 'Execute' to process the cur Press 'Save' to just change the pro Changed options will be effective v one-click 'Proc. Spectrum' button.	rent ( ocess when	dataset. sing options. pressing the	
Exponential Multiply (em)	•	LB [Hz] =	0.1
Fourier Transform (ft)			
Auto - Phasing (apk)			
Set Spectrum Reference (sref)			
Auto - Baseline Correction (abs)			
Plot (autoplot)		LAYOUT =	+/1D_H.xwp
Warn if processed data exist			

5. Click on Execute

6. Expand the spectrum from 5.5 ppm to 0 ppm





8. Adjust the **0** order phase on the peak at 5 ppm to phase the signal negative, which is

#### the selected exited peak

#### Figure 5.36



9. Click on 📙 to store the phase value

# 5.3.7 Plotting two spectra on to the same page

- 1. Display the selective NOESY spectrum
- 2. Click on it to enter the Multiple display option
- 3. Drag the Reference spectrum in to the spectral window





#### 4. Click on the 'Publish' tab in the TopSpin Menu bar

Figure	5 38
Figure	0.00

<u>S</u> tart	<u>A</u> cquire	Process	A <u>n</u> alyse	P <u>u</u> blish	<u>V</u> iew	<u>M</u> anage	0
		<u> </u>	opy 🗳 P <u>r</u> in	t 🗢 🖂 Plot	t Layout 🗟	P <u>D</u> F マ	- E-Mail

5. Click on the Sprint button to print the active window

# 5.3.8 Observations

# 5.4 1-D Selective TOCSY

#### 5.4.1 Introduction

Section 5.4 describes the acquisition and processing of a one-dimensional 1H selective gradient TOCSY experiment. The standard Bruker parameter set is **SELMLGP** and includes the pulse sequence **selmIgp** shown in Figure 5.16. It consists of the recycling delay, a radio-frequency (RF) pulse, a MLEV17 sequence for mixing and the acquisition time during which the signal is recorded.

Figure 5.39



#### 5.4.2 Reference spectrum

1. Run a **1D Proton** spectrum, following the instructions in **Chapter 3**, **1-D Proton** experiment, **3.2.2 Experiment setup**, through **3.2.4 Processing** 



Figure 5.40

# 5.4.3 Selective excitation region set up

#### 5.4.3.1 On resonance

NOTE: Make sure that the SW is large enough to cover the entire Spectrum accounting for the position of O1. The shaped pulse is applied on resonance (at the O1 position) The power level and width of the excitation pulse have to be known and entered into the Prosol parameter table

1. Type wrpa

Figure 5.41

Copy data set. I will be a 1-file d Please specify o	f NAME ends with ".top", the destina ataset (no expno/procno required). Jestination:	
NAME =	sel_tocsy	
EXPNO =	1	
PROCNO =	1	
DIP =	C:\data3.0	

- 2. Change NAME = sel\_tocsy
- 3. Click on OK
- 4. Type re

Figure 5.42

🚑 re	×
<ul> <li>Options</li> <li>O Display data in sa</li> <li>○ Display data in ne</li> </ul>	ime window w window
NAME =	sel_tocsy
EXPNO =	1
PROCNO =	1
DIR =	C:\data3.0
OK Cancel	Browse Find Help

- 5. Set Change NAME = sel\_tocsy
- 6. Click on OK
- 7. Expand peak at 4.8 ppm
- 8. Click on 🚺 to set the RF from cursor



9. Move the cursor line in to the center of the multiplet

10. Click the left mouse button to set the frequency

Figure 5.44



11. Click on 01

# 5.4.4 Setting up the Selective TOCSY

1. Click on the 'Start' tab in the TopSpin Menu bar

Figure 5.45							
<u>S</u> tart	<u>A</u> cquire <u>P</u> ro	cess A <u>n</u> alyse	P <u>u</u> blish	<u>V</u> iew	<u>M</u> anage	2	
	Create Data	set  Find Datas	et 🕥 Oper	n <u>D</u> ataset	🚺 Paste	Dataset 🔡	Read Pars.
2. Select 🔡	Read Pars.	by clicking or	n it				

#### Figure 5.46

📮 Parameter Sets: rpa	ar			
File Options He	elp	Source = C	:\Bruker\TopSpin3.0.b.42\	exp\stan\nmr\par 🛛 🖌
Find file names 🛛 🚽 e	enter any string, *, ?			
Class = Any	Dim = 🔄 🖸 Show	Recommended		
Type = 🔽 SubTyp	e = 😽 Reset Fil	ters		
SELCO1H	SELCOGP	SELGPSE	SELMLGP	SELMLZF1H
SELNO1H	SELNOGP	SELRO1H	SELROGP	SELU 13C
SELU 1H	SELU COSY	SELU HMBC	SELU HSQC	SELU ROESY
SELZG1H		_		
				Read Close

NOTE: Enter **SEL**\* in to the '**Find file names**' window and hit '**Enter**' to display all selective parameter sets shown in figure 5.48.

- 3. Select 'SELMLGP'
- 4. Click on Read...
- 5. Select the following parameter options: acqu, proc, outd
- 6. Click on the down arrow next to the 'Keep the following parameter' window
- 7. Select 'P1, O1, PLW1' from the pull down menu

Figure 5.47

😂 rpar	
Source Parameter Set = C:\Bruker\TopSpin Destination Data Set = selective_exp 2 1 1) Select the desired file types of the sourc 2) Press OK to copy them to the destinatio	13.0.b.42\exp\stan\nmr\par\SELCOC C:\data3.0 e parameter set n data set.
acqu	
proc	
outd	
Keep the following parameters:	
P 1. 01. PLW 1	~

- 8. Click on OK
- 9. Select the 'Title' tab by clicking on it
- 10. Make the following changes:

#### 1-D Selective gradient TOCSY experiment 30 mg Menthyl Anthranilate in DMSO-d6

- 11. Click on The to store the title
- 12. Select the 'AcquPars' tab by clicking on it

13. Click on the 'Aquire' tab in the TopSpin menu bar

Figure 5.48	
<u>Start</u> <u>A</u> cquire <u>P</u> roces	ss A <u>n</u> alyse P <u>u</u> blish <u>V</u> iew <u>M</u> anage 🕜
🕴 Sample 🗢 👫 Lock V T	une 🕶 🔱 Spin 👻 🛱 Shim 👻 🔏 Prosol 🚾 Gain 👻 🕨 Go 👻 Options 👻
Select 🦺 Spin 🚽 by clici	king on it
Figure 5.49	
	Turn sample rotation on (ro on)
	Turn sample rotation o <u>f</u> f (ro off)
	Change sample rotation rate (ro)
	MAS Pneumatic Unit (masdisp)

7. Select 'ro off' by clicking on it

NOTE: 1-D selective experiments should be run non spinning



NOTE: This will load the pulse width and power levels in to the parameter set.

# 5.4.5 Acquisition



# 5.4.6 Processing

1. Click on the 'Process' tab in the TopSpin Menu bar



- 3. Select 'Configure Standard Processing' by clicking on it
- 4. Diselect the following options:
  - 'Auto-Phasing (apk)' 'Set Spectrum Reference (sref)' 'Auto-Baseline correction (abs)' 'Warn if Processed data exist'



Press 'Execute' to process the cur Press 'Save' to just change the pro Changed options will be effective v one-click 'Proc. Spectrum' button.	rent ( ocess vhen	dataset. sing options. pressing the	
Exponential Multiply (em)		LB [Hz] =	0.1
Fourier Transform (ft)			
Auto - Phasing (apk)			
Set Spectrum Reference (sref)			
Auto - Baseline Correction (abs)			
Plot (autoplot)		LAYOUT =	+/1D_H.xwp

- 5. Click on Execute
- 6. Expand the spectrum fom 5.5 ppm to 0 ppm



# 7. Click on 🐴 Adjust Phase 😎

8. Phase all peaks positive





# 5.4.7 Plotting two spectra on to the same page

- 1. Display the selectice TOCSY spectrum
- 2. Click on it to enter the Multiple display option
- 3. Drag the Reference spectrum in to the spectral window







4. Click on the 'Publish' tab in the TopSpin Menu bar

Figure 5.56

<u>S</u> tart	<u>A</u> cquire	<u>P</u> rocess	A <u>n</u> alyse	P <u>u</u> blish	<u>V</u> iew	<u>M</u> anage	0
		<u><u> </u></u>	py 🗳 P <u>r</u> int	t 🗢 🕒 P <u>l</u> o	t Layout <del>v</del>	• ] 🦂 P <u>D</u> F マ	• <u>E</u> -Mail

5. Click on the rint vindow

# 5.4.8 Observations

#### B7169\_00\_01

# 6 1-D Carbon experiments

# 6.1 Sample

A sample of **30mg Brucine in CDCI3** is used for all experiments in this chapter

Figure 6.1



# 6.2 1-D Carbon Experiment

## 6.2.1 Introduction

Section 6.2 describes the acquisition and processing of a one-dimensional 13C NMR spectrum. The standard Bruker parameter set **C13CPD**, includes the pulse sequence **zgpg30**, shown in Figure 6.2. The 13C channel consists of the recycling delay, a RF pulse, and the acquisition time during which the signal is recorded. The pulse angle is shown to be 30 degrees. The two parameters, D1 and P1, correspond to the length of the recycle delay, and the length of the 90 degree RF pulse, respectively. The 1H channel consists of two decoupling pulses which can be power gated. The first pulse, an NOE build up pulse during the recycle delay may be of lower power then the second pulse on during the acquisition which is the true decoupling pulse. This can be useful to avoid RF heating on salty samples or probes where a higher decoupling power can be problematic.

Figure 6.2



The time intervals depicted in the pulse sequence diagrams are not drawn to scale. For example, d1 is typically a few seconds while p1 is typically a few microseconds in length.

# 6.2.2 Experiment set up

1. Click on the 'Start' tab in the TopSpin Menu bar

Figure 6.3

0	<u>S</u> tart	<u>A</u> cquire	Process	A <u>n</u> alyse	P <u>u</u> blish	<u>V</u> iew	<u>M</u> anage	2	
		Create	Dataset	📕 Find Dataset	t 🕥 Ope	n <u>D</u> ataset	Paste	Dataset	Read Pars.

2. Select Create Dataset by clicking on it

3. Enter the following information in to the 'New' window

Figure 6.4

NAME	Carbon_exp	5			
EXPNO	1				
PROCNO	1				
DIR	C:\data3	3.0			
Solvent				DMSO	
Experiment Dirs.	C:\Bruker\To	pSpin3	3.0.b.40\exp\	stan\nmr\par	
Experiment			C13CPD		
TITLE					
1- D 13C experi 30 mg Menthyl A	ment with 1H d Anthranilate in	lecoupl DMSO	ing -d6		

NOTE: The directory (DIR) is specific to how the data are stored and therefore may show different entries as the one in Figure 6.4 above. Click on the down arrow button to browse for a specific directory.

4. Click on OK

5. Select the 'AcquPars' tab by clicking on it

6. Make the following change

NS = 256

7. Click on the 'Aquire' tab in the TopSpin menu bar



#### 9. Select 'ej' by clicking on it

NOTE: Wait till the sample lift air is turned on and remove any sample which may have been in the magnet.



12. Select 'ij' by clicking on it

NOTE: Wait till the sample is lowered down in to the probe and the lift air is turned off. A licking sound may be heard.



Figure 6.8

💐 Solvents table	
△ Solvent	Description
Acetic	acetic acid-d4
Acetone	acetone-d6
Acetone_Hump	acetone-d6 (CHCI3)
C6D6	benzene-d6
CD2Cl2	methylenechloride-d2
CD3CN	acetonitrile-d3
CDCI3	chloroform-d
CDCI3_SENS	chloroform-d (ETB)
CH3CN+D2O	HPLC Solvent (Acetonitril/D2O)
D20	deuteriumoxide
DEE	diethylether-d10
Dioxane	dioxane-d8
DME	dimethylether-d6
DMF	dimethylformamide-d7
DMSO	dimethylsulfoxide-d6
EtOD	ethanol-d6
H2O+D2O	90%H2O and 10%D2O
Lock nucleus: 2H	OK Cancel

14. Select 'CDCI3' by clicking on it

15. Click on the down arrow V Tune -

NOTE: This performs a '**atma**' (automatic tuning) and requires a probe equipped with a automatic tuning module. Other options can be selected by clicking on the down arrow inside the '**Tune**' button.

17. Select	<b>掛</b> Sp <u>i</u> n <del>▼</del>	by clicking on it
Figure 6.9		
		Turn sample rotation on (ro on)
		Turn sample rotation off (ro off)
		Change sample rotation rate (ro)
		MAS Pneumatic Unit (masdisp)

18. Select 'ro on' by clicking on it

NOTE: Rotation may be turned off for probes such as BBI, TXI, TBI and for small sample probes.



NOTE: This will load the pulse width and power levels in to the parameter set.

### 6.2.3 Acquisition



### 6.2.4 Processing

1. Click on the 'Process' tab in the TopSpin Menu bar

Figure 6.10

		<u>S</u> tart	<u>A</u> cquire	Process	A <u>n</u> alyse	P <u>u</u> blish	<u>V</u> iew	<u>M</u> anage	2	
		∫ ∧ Pro <u>c</u> .	Spectrum <del>v</del>	Adjust P	hase 🗢 👌	Calib. A <u>x</u> is <del>⊲</del>	∙ XÅt P	ick P <u>e</u> aks <del>→</del>	∫ <u>I</u> ntegrate <del>→</del>	A <u>d</u> vanced <del>▼</del>
2. \$	Sele	ct 🔨	Proc S	nectrum	🚽 by c	lickina oı	n it			
			1 10 <u>0</u> . 0	pectrum	.,.					

NOTE: This executes a processing program including commands such as an exponential window function 'em', Fourier transformation 'ft', an automatic phase correction 'apk' and a baseline correction 'abs'. Other options are available by clicking on the down arrow inside the 'Proc. Spectrum' button.

3. Expand the spectrum to include all peaks



4. Select **Pick Peaks** by clicking on it

5. Click on 4 to define new peak picking range

6. Click the left mouse button and drag the cursor line from left to the right side of the spectrum



Figure 6.12

7. Click on manually adjust the minimum and maximum intensity levels

8. Click on the bottom line of the region box with the left mouse button and drag the line above the noise level, to set the minimum peak picking level

9. Click on the top line of the region box with the left mouse button and drag the line below unwanted peaks e.g. solvent peaks, to set the maximum peak picking level



10. Click on 🔄 to store the peak picking values





NOTE: To display the peak picking labels, right click inside the spectrum window and select '**Spectra Display Preferences**' by clicking on it. In the '**Spectrum components**' enable '**Peak labels**' and '**Peak annotations**'. Click '**Apply**' and click on '**Close**'

# 6.2.5 Plotting the 1D Carbon spectrum

- 1. Expand the spectrum (all peaks in display)
- 2. Click on the '**Publish**' tab in the TopSpin Menu bar

Figure 6.15

	,								
		<u>S</u> tart	<u>A</u> cquire	Process	A <u>n</u> alyse	P <u>u</u> blish	<u>V</u> iew	<u>M</u> anage	0
					py 🗳 P <u>r</u> in	t 🗢 🕒 P <u>l</u> ot	t Layout <del>~</del>		E-Mail
2.	Click o	n the d	own arrow	Plot	Layout <del>-</del>				

T Acquisition misned	. c./uataj.t	//carbon_exp/1/pda	100 U	V.	
Spectrum ProcPars	AcquPars	Title PulseProg P	eaks Integrals	Sample Structure Plo	Fid Acqu
🛈 🗇 Auto 🏡 🎴	+/1D_X.x	wp			
	<u></u>				
1- D 120	ovnovinont	with 10 decouplin	~		
30 mg Me	nthyl Anthi	anilate in DMSO-da	5		RRIKER
÷.	(e) (e)				
	h h	1 W L			Current Data Parameters NAME Carbon_exp EXPNO 1 FROCNO 1
					T3 - Augustifics Firstmater:           Data         2010751           Time         11,38           PROBED         2000751           Disc         2000751           PROBED         2000751           DOLVERT         2000           SS         128           DOLVERT         2000           SS         128           DR         16221,48           DR         16224,48           DR         177,733           DR         27,733           DR         27,733           DR         2,00000000 acc           DI1         2,0000000 acc           DI1         120
					PLN1 54.12799835 W SF01 75.4752949 MHz
	f .	ā č	1	L L L	CPDPRG2 waltz16 NUC2 18 PCPD2 95.00 use
		1			PLW2 22.65399933 W PLW12 0.30835000 W PLW13 0.24976000 W
					F2 - Processing parameters
Astronomic and provide	and the second second		and the second structures		SF 75.4677490 MHz WDW EM
. Lorison and	and the state of t	- 10 - 10 - 10 - 10 - 10 - 10 - 10 - 10		and the second secon	LB 1.00 Hz GB 0 PC 1.40
170 160	150 140	130 120 110 100	90 80 70 60	50 40 30 PE	m

NOTE: If desired, any changes can be administered using the Plot Editor tools.

4. Click on 'File' and select 'Print' by clicking on it

# 6.2.6 Observations

# 6.3 DEPT-135 Experiment

#### 6.3.1 Introduction

DEPT (Distortion less Enhancement by Polarization Transfer) is a polarization transfer technique used for the observation of nuclei with a small gyro magnetic ratio, which are J-coupled to 1H (most commonly 13C). DEPT is a spectral editing sequence, that is, it can be used to generate separate 13C sub spectra for methyl (CH3), methylene (CH2), and methine (CH) signals. DEPT makes use of the generation and manipulation of multiple quantum coherence to differentiate between the different types of 13C signals. Quaternary carbons are missing a direct bond proton, and as a result are absent from all DEPT spectra.

Section 6.3 describes the acquisition and processing of a one-dimensional 13C-DEPT135 NMR spectrum. The standard Bruker parameter set **C13DEPT135**, includes the pulse sequence **deptsp135**, shown in Figure 6.17. The 13C channel consists of the recycling delay, a 90 degree RF pulse, an editing delay D2 followed by a 180 degree RF pulse and the acquisition time during which the signal is recorded. The editing delay D2 is 1/2\*J(XH). The 1H channel consists of two three pulses, a 90 degree, a 180 degree, followed by a 135 degree RF pulse and are separated by the editing delay D2. The final 135 degree 1H pulse selects the CH3, CH2 or CH signals. The protons are decoupled during the acquisition period.





The time intervals depicted in the pulse sequence diagrams are not drawn to scale. For example, d1 is typically a few seconds while p1 is typically a few microseconds in length.

## 6.3.2 Experiment set up

NOTE: This experiment usually follows a regular 1H decoupled 13C experiment. The result of a DEPT-135 experiment shows only the protonated carbons with the CH and CH3 as positive and the CH2 as negative signals.

1. Click on the 'Start' tab in the TopSpin Menu bar

Ctart Aan	uiro Drosoco	Analyza		Managa	0
	Create Dataset	Find Dataset	Puplish view	et <b>F</b> Past	e Dataset 📓 Read Pars.
	Torre Darreet)		- open <u>D</u> ata		
2. Select Crea	te Dataset	by clicking	on it		
3 Enter the followin	a information	in to the '	New' window		
	ig information			v	
Figure 6.19					
	🚔 New				
	For multi-receiver Please define the	parameters acco experiments sevi number of receiv	irding to the selecti eral datasets are ci ers in the box belo	ed experiment eated. v.	туре.
	NAME	Carbon_exp			
	EXPNO	3			
	PROCNO	1			
	DIR	C:\data3.0			*
	Solvent	1	[	MSO	<b>*</b>
	Experiment Dirs.	C:\Bruker\TopS	pin3.0.b.40\exp\sta	n\nmr\par	<b>*</b>
	Experiment		C13DEPT90		
		10 ovporiment			
	30 mg Menthyl A	nthranilate in DN	ISO-d6		
	Show new dat	aset in new wind	0\\/		
	1 Receivers (	(1,2,16)			
		ОК	Cancel More	Info He	elp
l					

NOTE: The directory (DIR) is specific to how the data are stored and therefore may show different entries as the one in Figure 6.4 above. Click on the down arrow button to browse for a specific directory.

- 4. Click on OK
- 5. Select the 'AcquPars' tab by clicking on it
- 6. Make the following change

#### NS = **128**

7.Click on the 'Aquire' tab in the TopSpin menu bar

Figu	re 6.	20								
		<u>S</u> tart	<u>A</u> cquire	Process	A <u>n</u> alyse P	ublish <u>V</u> ie	w <u>M</u> anag	e 🕜		
		🕴 Sample	• <b>▼</b> <u>₩ L</u> oc	ck V T <u>u</u> ne <del>√</del>	Sp <u>i</u> n <del>▼</del>	Shim <del>→</del>	Prosol	<u></u>	▶ Go 🗢	Options 🗢
		(	_							

8. Select **A Prosol** by clicking on it

NOTE: This will load the pulse width and power levels in to the parameter set.

# 6.3.3 Acquisition 1. Select Gain by clicking on it

NOTE: To adjust rg manually, click on the down arrow inside the 'Gain' icon



# 6.3.4 Processing

1. Click on the 'Process' tab in the TopSpin Menu bar



NOTE: This executes a processing program including commands such as an exponential window function 'em', Fourier transformation 'ft', an automatic phase correction 'apk' and a baseline correction 'abs'. Other options are available by clicking on the down arrow inside the 'Proc. Spectrum' button. Do to the fact that a DEPT135 spectrum contains negative and positive peaks, there is the possibility of getting phase results that are 180 degrees off. In this case, click on the 'Adjust Phase' button to enter the manual phase routine and reverse the spectrum by clicking on the '180' icon.



# 6.3.5 Observations

# 6.4 DEPT-90 Experiment

#### 6.4.1 Introduction

Section 6.4 describes the acquisition and processing of a one-dimensional 13C-DEPT90 NMR spectrum. The standard Bruker parameter set **C13DEPT90**, includes the pulse sequence **dept90**, shown in Figure 6.23. The 13C channel consists of the recycling delay, a 90 degree RF pulse, an editing delay D2 followed by a 180 degree RF pulse and the acquisition time during which the signal is recorded. The editing delay D2 is 1/2\*J(XH). The 1H channel consists of three pulses, a 90 degree, a 180 degree, followed by a 90 degree RF pulse and are separated by the editing delay D2. The final 90 degree 1H pulse selects the CH signals only. The protons are decoupled during the acquisition period.

Figure 6.23



The time intervals depicted in the pulse sequence diagrams are not drawn to scale. For example, d1 is typically a few seconds while p1 is typically a few microseconds in length.

## 6.4.2 Experiment set up

NOTE: The DEPT90 experiment usually follows a regular 1H decoupled 13C experiment and a DEPT-135 experiment. It is used to assign the methine (CH) signals.

1. Click on the 'Start' tab in the TopSpin Menu bar

Figure 6.24

<u>S</u> tart	<u>A</u> cquire	Process	A <u>n</u> alyse	P <u>u</u> blish	<u>V</u> iew	<u>M</u> anage	0	
	C <u>r</u> eate	Dataset	📕 Find Dataset	t 🔄 Ope	n <u>D</u> ataset	Paste	Dataset	Read Pars.

- 2. Select Create Dataset by clicking on it
- 3. Enter the following information in to the 'New' window

Figure 6.25

NAME	Carbon_exp					
EXPNO	2					
PROCNO	1					
DIR	C:\data3.0			1		
Solvent	147		DMSO	1		
Experiment Dirs.	C:\Bruker\TopS	Spin3.0.b.40\	exp\stan\nmr\par	•		
Experiment		C13DEPT135				
TITLE						
1- D 13C DEPT1 30 mg Menthyl A	35 experiment nthranilate in DN aset in new wind	1SO-d6 ow				

NOTE: The directory (DIR) is specific to how the data are stored and therefore may show different entries as the one in Figure 6.4 above. Click on the down arrow button to browse for a specific directory.

- 4. Click on OK
- 5. Select the 'AcquPars' tab by clicking on it
- 6. Make the following change
- NS = 128

7.Click on the 'Aquire' tab in the TopSpin menu bar



NOTE: This will load the pulse width and power levels in to the parameter set.

# 6.4.3 Acquisition

1. Select <u>🚾 G</u>ain 🤜 by clicking on it

NOTE: To adjust rg manually, click on the down arrow inside the 'Gain' icon


#### 6.4.4 Processing

1. Click on the 'Process' tab in the TopSpin Menu bar

Figu	ure 6.	27								
		<u>S</u> ta	rt <u>A</u> cquire	Process	A <u>n</u> alyse	P <u>u</u> blish	<u>V</u> iew	<u>M</u> anage	0	
		A Pro	<u>c</u> . Spectrum <del>v</del>	Adjust P	Phase 🗢 📝 🎊	Calib. A <u>x</u> is	✓ <sup>*</sup> P	ick P <u>e</u> aks <del>→</del>	∫ <u>I</u> ntegrate <del>→</del>	A <u>d</u> vanced <del>▼</del>
2. \$	Sele	ct 🧾	Pro <u>c</u> . S	pectrum	🤟 by c	licking o	n it			

NOTE: This executes a processing program including commands such as an exponential window function 'em', Fourier transformation 'ft', an automatic phase correction 'apk' and a baseline correction 'abs'. Other options are available by clicking on the down arrow inside the 'Proc. Spectrum' button.

#### Figure 6.28



# 6.4.5 Observations

# 7 2-D Heteronuclear experiments

# 7.1 Sample

A sample of **30mg Menthyl Anthranilate in DMSO-d6** is used for all experiments in this chapter

Figure 7.1



# 7.2 2D edited HSQC

#### 7.2.1 Introduction

The **HSQC** (Heteronuclear Single Quantum Coherence) experiment performs and H,C-correlation via the 13C chemical shift evolution of the double-quantum coherence. This method is superior to other heteronuclear experiments in the case of a crowded 13C NMR spectrum. In the sequence shown in Figure 7.2., the signals are not broadened by homonuclear H,H coupling in F1. It is possible to obtain a complete editing of inverse -recorded 1-D H,X correlation spectra. This kind of multiplicity determination has been achieved by including an editing period within HSQC, abbreviated as E-HSQC. In the experiment shown here the standard Bruker parameter set **HSQCEDETGP** is used and the graphical display of the pulse program **hsqcedetgp** is shown in Figure 7.2.



The time intervals depicted in the pulse sequence diagrams are not drawn to scale. For example, d1 is typically a few seconds while p1 is typically a few microseconds in length.

#### 7.2.2 Preparation experiment

1. Run a 1D Proton spectrum, following the instructions in Chapter3, 1-D Proton experiment, Paragraph 3.2.2 Experiment setup through 3.2.4 Processing.



Figure 7.3

#### 7.2.3 Setting up the HSQC experiment

1. Click on the 'Start' tab in the TopSpin Menu bar	
Figure 7.4	
Start Acquire Process Analyse Publish View Manage	
🗋 C <u>r</u> eate Dataset 📓 F <u>i</u> nd Dataset 🖄 Open <u>D</u> ataset 📭 Paste Dataset 📄 Read Pars.	
<ol> <li>Select Create Dataset by clicking on it</li> <li>Enter the following information in to the 'New' window</li> </ol>	
Figure 7.5	
🔄 New 🔀	
Prepare for a new experiment by creating a new data set and initializing its NMR parameters according to the selected experiment type. For multi-receiver experiments several datasets are created. Please define the number of receivers in the box below.	
NAME Inverse_exp	
EXPNO 2	
PROCNO 1	
Experiment Dirs C:\Bruker\TopSpin3.0.b.40\exp\stan\nmr\par	
Experiment HSQCEDETGP	
2-D edited HSQC experiment 30 mg Menthyl Anthranilate in DMSO-d6  Show new dataset in new window  Receivers (1,2,16)	
OK Cancel More Info Help	

NOTE: The directory (DIR) is specific to how the data are stored and therefore may show different entries as the one in Figure 4.5 above. Click on the down arrow button to browse for a specific directory.

4. Click on OK

5. Click on the 'Aquire' tab in the TopSpin menu bar

Figure 7.6 0 Start Acquire Process A<u>n</u>alyse P<u>u</u>blish View <u>M</u>anage 🌵 Sample 👻 🌞 Lock 🔰 Tune 💌 🔱 Spin 🔻 🛱 Shim 👻 🛃 Prosol 🔲 SetLimits 💹 Gain 💌 👂 Go 💌 Options 🗢 6. Select J Spin ▼ by clicking on it

	Turn sample rotation on (ro on)	
	Turn sample rotation off (ro off)	
	Change sample rotation rate (ro)	
	MAS Pneumatic Unit (masdisp)	
		'
7. Select 'ro off' by clicking of	on it	

NOTE: 2-D experiments should be run non spinning 8. Select **Prosol** by clicking on it

NOTE: This will load the pulse width and power levels in to the parameter set.



10. To open the 1D Proton spectrum, right click on the dataset name in the browser window (e.g. **Inverse\_exp 1**) and select 'Display' or click and hold the left mouse button for dragging the 1D Proton dataset in to the spectrum window

11. Expand the spectrum to display all peaks, leaving ca. 0.2 ppm of baseline on either side of the spectrum

NOTE: The solvent peak may be excluded if it falls outside of the region of interest. Digital filtering however is only applied in F2 and the solvent peak is folding in F1.



NOTE: The display changes back to the 2D data set. The parameter set **HSQCEDETGP** has a fixed F1 sweep width of 160 ppm and it is big enough to cover the protonated resonances for a broad range of samples. If desired, changes to the F1 sweep width can be done by using the '**Set\_limits**' button for a second time. In this case a 1-D **C13DEPT45** or **C13DEPT135** experiment on the same sample has to be observed. As an example to set the F1 limit, follow the steps below.





Figure 7.11

14. To open the 1D C13DEPT spectrum, right click on the dataset name in the browser window (e.g. **Carbon\_exp 2**) and select 'Display' or click and hold the left mouse button for dragging the 1D C13DEPT dataset in to the spectrum window

15. Expand the spectrum to display all peaks, leaving ca. 2 ppm of baseline on either side of the spectrum

NOTE: The solvent peak may be excluded if it falls outside of the region of interest. Digital filtering however is only applied in F2 and the solvent peak is folding in F1.



	Figure 7.13	
	د د	X 13C spectral limits copied for F1 dimension. SW: 132.9571 ppm centered at 76.350 ppm SR: 37.73 Hz
		Close
	17. Click on Close	
7 2 1	Acquisition	

#### 7.2.4 Acquisition



# 7.2.5 Processing

1. Click on the 'Process' tab in the TopSpin Menu bar



NOTE: This executes a standard processing program **proc2**. To configure this program or select the right options, click on the down arrow inside the '**Proc. Spectrum**' button. Since this is a phase sensitive experiment the phase correction **apk2d** have to be enabled.



# 7.2.6 Plotting

- 1. Use the 🦣 buttons to adjust for a suitable contour level
- 2. Click on the 'Publish' tab in the TopSpin Menu bar

Figure 7.16

	<u>S</u> tart	<u>A</u> cquire	Process	A <u>n</u> alyse	P <u>u</u> blish	<u>V</u> iew	<u>M</u> anage	0
-				py 🗳 P <u>r</u> int	▼ Plot	Layout <del>v</del>		E-Mail







NOTE: This will print the active window with the colors displayed in the TopSpin window. Using the '**plot**' option starting the plot editor, the default layout is designed not to show the F1 projection (see Figure 7.18 below. A new layout has to be created to add the F1 projection.





# 7.2.7 Observations

# 7.3 2D HMBC experiment

#### 7.3.1 Introduction

HMBC (Heteronuclear Multiple Bond Correlation) spectroscopy is a modified version of HMQC suitable for determining long-range 1H-13C connectivity. Since it is a long-range chemical shift correlation experiment the pulse program contains a low pass filter to suppress the one bond correlation and is a gradient-selected version which is not phase-sensitive. The experiment is performed without 13C decoupling to distinguish signals coming from the one bond coupling and the standard Bruker parameter set **HMBCGP** is used. The graphical display of the pulse program **hmbcgplpndqf** is shown in Figure 7.7

hmbcgplpndqf

Figure 7.19



The time intervals depicted in the pulse sequence diagrams are not drawn to scale. For example, d1 is typically a few seconds while p1 is typically a few microseconds in length.

#### 7.3.2 Preparation experiment

1. Run a 1D Proton spectrum, following the instructions in Chapter3, 1-D Proton experiment, Paragraph 3.2.2 Experiment setup through 3.2.4 Processing.



# 7.3.3 Setting up the HMBC experiment

1. Click on the 'Start' tab in the TopSpin Menu bar

Figure 7.21

	<u>S</u> tart	<u>A</u> cquire	Process	A <u>n</u> alyse	P <u>u</u> blish	<u>V</u> iew	<u>M</u> anage	0	
		Create	Dataset	📕 Find Datase	t 🕥 Ope	n <u>D</u> ataset	Paste	Dataset	Read Pars.

2. Select Create Dataset by clicking on it

3. Enter the following information in to the 'New' window

Figure	7.22
riguic	1.22

NAME	Inverse_6	ехр		
EXPNO	3			
PROCNO	1			
DIR	C:\da	ta3.0		~
Solvent			DMSO	1
Experiment Dirs.	C:\Bruker	TopSpin3.0.b.40	)\exp\stan\nmr\par	*
Experiment		НМВСС	ЭР	1
TITLE 2-D edited HSG	C experimer	nt In DMSO de		

NOTE: The directory (DIR) is specific to how the data are stored and therefore may show different entries as the one in Figure 4.5 above. Click on the down arrow button to browse for a specific directory.

- 4. Click on OK
- 5. Click on the 'Aquire' tab in the TopSpin menu bar





NOTE: 2-D experiments should be run non spinning

8. Select Frosol by clicking on it



10. To open the 1D Proton spectrum, right click on the dataset name in the browser window (e.g. **Inverse\_exp 1**) and select 'Display' or click and hold the left mouse button for dragging the 1D Proton dataset in to the spectrum window

11. Expand the spectrum to display all peaks, leaving ca. 0.2 ppm of baseline on either side of the spectrum

NOTE: The solvent peak may be excluded if it falls outside of the region of interest. Digital filtering however is only applied in F2 and the solvent peak is folding in F1.



12. Click on <b>OK</b> to assign the new limit
Figure 7.27
1H spectral limits copied for F2 dimension. SW: 8.4682 ppm O1P: 4.234 ppm
Close
13. Click on Close

NOTE: The display changes back to the 2D data set. The parameter set **HMBCGP** has a fixed F1 sweep width of 222 ppm and it is big enough to cover all Carbon resonances for a broad range of samples. If desired, changes to the F1 sweep width can be done by using the '**Set\_limits**' button for a second time. In this case a 1-D **C13CPD** experiment on the same sample has to be observed. As an example to set the F1 limit, follow the steps below.

Select	Se <u>t</u> Limits	by clicking on it
Figure 7.	28	
	🔄 setli	imits
	0	<ul><li>Close this dialog box after setting frequencies.</li><li>1. Open 1D dataset from Browser.</li><li>2. Zoom into region of interest.</li><li>3. Click OK to set frequencies and return to original dataset.</li></ul>
		OK Cancel

14. To open the 1D C13DEPT spectrum, right click on the dataset name in the browser window (e.g. **Carbon\_exp 1**) and select 'Display' or click and hold the left mouse button for dragging the 1D C13DEPT dataset in to the spectrum window

15. Expand the spectrum to display all peaks, leaving ca. 2 ppm of baseline on either side of the spectrum

NOTE: The solvent peak may be excluded if it falls outside of the region of interest. Digital filtering however is only applied in F2 and the solvent peak is folding in F1.





# 7.3.5 Processing

1. Click on the '**Process**' tab in the TopSpin Menu bar



NOTE: This executes a standard processing program **proc2**. The message shown in Figure 7.31 pops up in case of a magnitude 2D experiment and the apk2d option is enabled. To configure the processing program follow the steps below.

3. Click on the down arrow i	nside the 🥂 Proc. Spectrum
Figure 7.33	
	Configure Standard Processing (proc2d)
	Process F2+F1 Axis (xfb)
	Process Only F2 Axis (xf2)
	Process Only F1 Axis (xf1)
	Symmetrize Spectrum (sym)
	Start Automation AU Program (xaup)

3. Select 'Configure Standard Processing' by clicking on it



	enecco	
Press 'Execute' to process the current of Press 'Save' to just change the process Changed options will be effective when p one-click 'Proc. Spectrum' button.	atase ng o press	et. ptions. ing the
Fourier Transform (xfb)	$\overline{\mathbf{v}}$	
Auto - Phasing (apk2d)		
Auto - Baseline Correction [F2] (abs2)		
Auto - Baseline Correction [F1] (abs1)	<b>V</b>	
Plot (autoplot)		LAYOUT = +/2D_hom.xwp
Warn if processed data exist		
		Save Execute Cancel

NOTE: To avoid the message shown in Figure 7.31 the option 'Auto-Phasing (apk2d)' may be disabled for magnitude like 2D experiment.



# 7.3.6 Plotting

- 1. Use the the buttons to adjust for a suitable contour level
- 2. Click on the 'Publish' tab in the TopSpin Menu bar



NOTE: This will print the active window with the colors displayed in the TopSpin window. Using the '**plot**' option starting the plot editor, the default layout is designed not to show the F1 projection (see Figure 7.37 below. A new layout has to be created to add the F1 projection.

#### Figure 7.38



# 7.3.7 Observations

#### B7169\_00\_01

# 8 Determination of 90 degree pulses

# 8.1 Introduction

This chapter describes pulse calibration procedures for 1H and 13C. It is assumed that the user is already familiar with acquisition and processing of simple 1D NMR spectra. Chapter 3 (1-D Proton experiment) and chapter 6 (1-D Carbon experiments).

NOTE: This chapter is intended as a guide for calibrating the 90 degree pulse of a probe or verifying the values observed using ATP.

# 8.2 Proton 90 degree transmitter pulse

Standard Test Sample:

0.1% Ethylbenzene in CDCI3

#### 8.2.1 Parameter setup

1. Click on the 'Start' tab in the TopSpin Menu bar

<u>S</u> tart <u>A</u> cquire	Process Analyse	P <u>u</u> blish <u>V</u> iew	Manage 🕜	
C <u>r</u> eate E	Dataset [ 😹 F <u>i</u> nd Dataset	🕥 Open <u>D</u> ataset	Paste Dataset	Read Pars.

2. Select Create Dataset by clicking on it

3. Enter the following information in to the 'New' window

Figure	82
rigule	0.2

NAME	proton_90			
EXPNO	1			
PROCNO	1			
DIR	C:\data	3.0		-
Solvent			CDCI3	-
Experiment Dirs.	C:\Bruker\T	opSpin3.0.b.4	3\exp\stan\nmr\par	•
Experiment		PROTO	N	1
90 degree pulse 0.1% Ethylbenze	e test for Prote ene in CDCl3	on		

NOTE: The directory (DIR) is specific to how the data are stored and therefore may show different entries as the one in Figure 8.2 above. Click on the down arrow button to browse for a specific directory.

4. Click on OK

5. Run a **1D Proton** spectrum, following the instructions in **Chapter 3**, **1-D Proton experiment, Paragraph 3.2.2 Experiment setup, step 5** through **3.2.4 Processing**.

Figure 8.3



6. Expand peak at 2.7 ppm



- 8. Move the cursor line in to the center of the multiplet
- 9. Click the left mouse button to set the frequency

Figure 8.5



- 10. Click on O1
- 11. Select the 'AcquPars' tab by clicking on it
- 12. Make the following changes:

PULPROG = zg TD = 4048 SW [Hz] =300 D1 [sec] = 30 DS = 0 NS = 1

- 13. Select the 'ProcPars' tab by clicking on it
- 14 Make the following changes:

SI = **2024** LB [Hz] = **1** PH\_mod = select **'pk**'

15. Click on the '**Aquire**' tab in the TopSpin menu bar

Figure 8.	6									
	<u>S</u> tart	Acquire	Process	A <u>n</u> alyse	P <u>u</u> blish	<u>V</u> iew	Manag	ge 🕜		
	🌵 Sample 🚽	• # <u>L</u> ock	√ V T <u>u</u> ne •	🗸 🕹 Sp <u>i</u> r	n <del>⊽</del> िन Shi	m <b>→</b> 🔏	Pr <u>o</u> sol	<u>I∽</u> <u>G</u> ain <del>▼</del>	Þ Go 🗢	Options 🗢
Select		n 🚽 by	y clicking	on it						
	A obs		,	•						
Figure 8.	7									
			TU	i <u>r</u> n samp	le rotation	n on (ro	on)			
			Т	irn samp	le rotatior	n o <u>f</u> f (ro	off)			
			<u>C</u> ł	nange sa	mple rota	tion rat	e (ro)			
			M	AS Pneu	matic Unit	(masdi	sp)			
			L				100.00			

16. Select '**ro off**' by clicking on it

NOTE: This test should be run non spinning

# 8.2.2 Acquisition

1. Select	<u>I∽ G</u> ain <del>▼</del>	by clicking on it
2. Select	▶ Go 🗸	by clicking on it

#### 8.2.3 Processing

1. Click on the 'Process' tab in the TopSpin Menu bar

Figure 8.8

		<u>S</u> tart	<u>A</u> cquire	Process	A <u>n</u> alyse	P <u>u</u> blish	<u>V</u> iew	<u>M</u> anage	0	
		Λ Pro <u>c</u> . 8	Spectrum <del>v</del>	Adjust P	Phase 🗢 🔥	Calib. A <u>x</u> is <del>-</del>	N Pie	ck P <u>e</u> aks <del>⊸</del>	∫ Integrate <del>→</del>	A <u>d</u> vanced <del>▼</del>
2.	Click	c on the	down a	rrow insid	de the	A Pro <u>c</u> . S	Spectru	um 🚽 bi	utton	

Configure Standard Processing (proc1d)
Window Multiplication (wm)
Fourier <u>T</u> ransform (ft)
Start Automation AU Program (xaup)

- 3. Select 'Configure Standard Processing' by clicking on it
- 4. Deselect the following options:

'Set Spectrum Reference (sref)' 'Auto-Baseline correction (abs)' 'Warn if Processed data exist'

Figure 8.10

Press 'Execute' to process the cur Press 'Save' to just change the pro Changed options will be effective v one-click 'Proc. Spectrum' button.	rent oces: when	dataset. sing options. pressing the	
Exponential Multiply (em)		LB [Hz] =	1
Fourier Transform (ft)			
Auto - Phasing (apk)			
Set Spectrum Reference (sref)			
Auto - Baseline Correction (abs)			
Plot (autoplot)		LAYOUT =	+/1D_H.xwp

5. Click on Execute

6. Expand the spectrum form 2.8 ppm to 2.5 ppm



- 7. Click on the right mouse button inside the spectral window
- Figure 8.12



- 8. Select 'Save Display Region to ... ' by clicking on it
- Figure 8.13



#### 9. Enable 'Parameters F1/2'

- 10. Click on OK
- 11. Type wpar to store the parameter for future use
- 12. Select the user parameter directory

Figure 8.14

Source = C:\Bruker\TopSpin3.0.b.43\exp\stan\nmr\par\user v

#### Figure 8.15

Find file names	enter any string * ?			
Diass = M Dim =	Show Recon	nmended		
Type = 🛛 🖌 SubTy	pe = 🛛 🖌 🛛 Reset Fi	Iters		
411.050				
C13DEPT135p mod	COSVGP fixsw	E10 mod	E19COSVGP test	H1n00 urea
HCCHCOGP3D pz	HMBCEDETGPI3ND	HSOCEDETGP mod	HSOCETGPN15	MI EVETGPSW
NP 1H	NP C13CPD	NP COSY	NP DEPTQ	NP HMBC
NP HSQC	NP JRES	NP ZG30	PRO128PP	PROTON 3exp
ROESYETGPSW	SELCOGP.pz	SELMLGP.pz	SELNOGP.mod	SELNOGP.pz
SELROGP.pz	SI29IGSW	SOLVSUP WET		

13. Click on Write New...

Figure 8.16



14. Type proton\_90 in to the new name window

Figure 8.17

Source Data Set	= proton_90 1 1 C:\data3.0
1) Select the des	sired file types of the source data set
2) Press OK to c	opy them to the destination parameter set.
acqu	
proc	
outa	
title	
Destination Dir =	C:\Bruker\TopSpin3.0.b.43\exp\stan\nmr\par\user
Destination Dir =	C:\Bruker\TopSpin3.0.b.43\exp\stan\nmr\par\user

- 16. Select all parameter options
- 17. Click on OK

#### Figure 8.18

	101): 			
File Options He	lp	Source = C:\Brui	ker\TopSpin3.0.b.43\e	xp\stan\nmr\par\user
Find file names 🛛 💆 e	nter any string, *, ?			
Class = 🔽 Dim =	Show Recom	imended		
Гуре = 🔄 SubTyp	ie = 🔽 Reset Fil	Iters		
1H 256	C13CPD128.mod	C13CPD2K	C13CPD64	C13DEPT135NS32
C13DEPT135p.mod	COSYGP.fixsw	F19_mod	F19COSYGP.test	H1p90_urea
HCCHCOGP3D.pz	HMBCEDETGPI3ND	HSQCEDETGP.mod	HSQCETGPN15	MLEVETGPSW
NP_1H	NP_C13CPD	NP_COSY	NP_DEPTQ	NP_HMBC
NP_HSQC	NP_JRES	NP_ZG30	PRO128PP	PROTON_3exp
proton 90	ROESYETGPSW	SELCOGP.pz	SELMLGP.pz	SELNOGP.mod
SELNOGP.pz	SELROGP.pz	SI29IGSW	SOLVSUP WET	

18. Click on Close

# 8.2.4 Determine the 90<sup>0</sup> pulse

1. Click on the 'Aquire' tab in the TopSpin menu bar

Figure 8.19

	<u>S</u> tart	Acquire	Process /	A <u>n</u> alyse P	ublish <u>V</u> i	ew <u>M</u> ana	ige 🕜			
	🕴 Sample	e <del>▼</del>   <b>‡</b> oc	k	<mark>∛</mark> Sp <u>i</u> n <del>⊽</del>	🖙 Shim 🗢	Prosol	<u> </u>	▶ Go 🗢	Options 🗢	



- 3. Select 'Optimize Acquisition Params (popt)' by clicking on it
- 4. Make the following changes:

OPTIMIZE = Step by step PARAMETER = p1 OPTIMUM = POSMAX STARTVAL = 2 NEXP = 20 VARMOD = LIN INC = 2

#### Figure 8.21

store as 2D data	(ser file)							
The AU program	specified	t in AUNM will b	e executed		WDW= EM			
Perform automat	tic baselin	e correction (A	BSF)		PH_mod= no	)		
Overwrite existin	g files (di	sable confirmat	ion Message)		FT mod= fs			
Stop sample spir	nning at th	he end of optim	ization (mash)					
Run optimization	in backg	round						
OPTIMIZE	GROUP	PARAMET	OPTIMUM	STARTVAL	ENDVAL	NEXP	VARMOD	INC
Step by step		p1	POSMAX	2		20	LIN	2
Start optimize	Skip curi	rent opti	now protocol	Add parame.	. Rest	ore )	Save )[	Read array f

5. Click on Save



6. Click on Start optimize

Figure 8.22

Number of experiments: 20	)
total experiment time will b Continue ? [y   n]	e: 12 min 0 sec

- 7. Enter y in to the poptau window
- 8. Click on OK

NOTE: The parameter optimization starts. The spectrometer acquires and processes 20 spectra with incrementing the parameter p1 from 2 usec by 2 usec to a final value of 40 usec. For each of the 20 spectra, only the spectral region defined above is plotted, and all the spectra are plotted side-by-side in the file proton\_90/1/999 as shown in Figure 8.18.





NOTE: The POSMAX value of **p1** is displayed in the title window which is the 90 degree pulse, along with the experiment number and the NEXP value. Write this value down. To obtain a more accurate 90 degree pulse measurement, follow the steps below.

9. Close the popt setup window

- 10. Type re 1 1
- 11. Type p1

12. Enter the value which corresponds to a 360 degree pulse (four times the POSMAX value)

- 13. Type zg
- 14. Type efp

15. Change p1 slightly and repeat steps 12 and 13, until the quartet undergoes a zero crossing as expected for an exact 360 degree pulse.

NOTE: The quartet signal is negative for a pulse angle slightly less then 360 degree and positive when the pulse angle is slightly more then 360 degree.

16. Simply divide the determine 360 degree pulse value by 4. This will be the exact 90 degree pulse length for the proton transmitter on the current probe

# 8.2.5 Observations
#### 8.3 Carbon 90 degree transmitter pulse

Standard Test Sample:

ASTM (60% C6D6 / 40% p-Dioxane)

#### 8.3.1 Parameter setup

1. Click on the 'Start' tab in the TopSpin Menu bar

Figure 8.24									
Sta	art	<u>A</u> cquire	Process	A <u>n</u> alyse	P <u>u</u> blish	<u>V</u> iew	<u>M</u> anage	2	
		C <u>r</u> eate	Dataset	📕 F <u>i</u> nd Dataset	t) 🗐 Oper	n <u>D</u> ataset	Paste	Dataset	Read Pars.
2. Select	C <u>I</u> follo	<mark>eate D</mark> wing inf	ataset formation	by clicking n in to the '	ı on it New' w	indow			
Figure 8.25									
0		🔄 Ne	w						
		Prepa initial For n Pleas	are for a new izing its NMR nulti-receiver se define the	experiment by c parameters acco experiments sev number of receiv	reating a ne ording to the eral dataset vers in the b	w data set e selected e is are creat ox below.	and experiment ty ed.	pe.	
		NAM	IE	carbon_90					
		EXP	NO	1					
		PRC	OCNO	1					
		DIR		C:\data3.0				~	
		Solv	ent	-		C6D	6	~	
		Expe	eriment Dirs.	C:\Bruker\TopS	Spin3.0.b.43	\exp\stan\n	mr\par	~	
		Expe	eriment		C13CPD	2		*	
		TITL	.E						
		90 AS	degree pulse FM (60% C6E	test for 13C 06 / 40% Dioxane	e)				

NOTE: The directory (DIR) is specific to how the data are stored and therefore may show different entries as the one in Figure 8.25 above. Click on the down arrow button to browse for a specific directory.

Cancel

More Info.

Help

Show new dataset in new window Receivers (1,2, ...16)

OK

1

4. Click on OK

5. Run a 1D Carbon spectrum, following the instructions in Chapter 6, 1-D Carbon experiments, Paragraph 6.2.2 Experiment setup, step 5 making the following acquisition parameter changes:

PULPROG = zg

DS = 0 NS = 1

6. Continue with 6.2.4 Processing.



- 7. Expand peak at 67 ppm
- 8. Click on 5 to set the RF from cursor



9. Click the left mouse button to set the frequency

Figure 8.28

🤤 01/02/03	X
Define SFO1/O1 1	frequencies
SFO1 [MHz] =	75.472790
O1/2/3 [Hz] =	5041.45
01 02	O3 Cancel

10. Click on O1

11. Select the 'AcquPars' tab by clicking on it

12. Make the following changes:

TD = **4048** SW [Hz] =**20** D1 [sec] = **60** 

13. Select the 'ProcPars' tab by clicking on it

14 Make the following changes:

SI = 2024 LB [Hz] = 3.5

PH\_mod = select '**pk**'

15. Click on the 'Aquire' tab in the TopSpin menu bar

Figure 8.29	
<u>Start</u> <u>A</u> cquire <u>P</u> roces	s A <u>n</u> alyse P <u>u</u> blish <u>V</u> iew <u>M</u> anage 🚱
🕴 Sample 🚽 🏥 Lock	une 🔻 🔱 Spin 👻 🛱 Shim 👻 🕂 Prosol 💹 Gain 👻 Þ Go 👻 Options 👻
Select <mark>↓ Spin ↓</mark> by click Figure 8.30	king on it
	Tu <u>r</u> n sample rotation on (ro on)
	Turn sample rotation off (ro off)
	Change sample rotation rate (ro)
	MAS Pneumatic Unit (masdisp)

16. Select 'ro off' by clicking on it

NOTE: This test should be run non spinning

### 8.3.2 Acquisition

1. Select	<u>I∽ Gain</u> <del>▼</del>	by clicking on it
2. Select	B Go マ	by clicking on it

### 8.3.3 Processing

1. Click on the 'Process' tab in the TopSpin Menu bar

Figure 8.31



- 3. Select 'Configure Standard Processing' by clicking on it
- 4. Deselect the following options:
  - 'Set Spectrum Reference (sref)'

### 'Auto-Baseline correction (abs)' 'Warn if Processed data exist'

Figure 8.33



- 5. Click on Execute
- 6. Expand the spectrum from 71 ppm to 63 ppm

#### Figure 8.34



7. Click on the right mouse button inside the spectral window

Figure 8.35

Togale S	pectrum Overview
Show Fu	Il Spectrum
Toggle P	arameter <u>W</u> indow
Spe <u>c</u> tra I	Display Preferences
Save Dis	play Regi <u>o</u> n To
Restore I	Display Region From Params. F1/2
Set Plot I	Height At Specific Cursor Position
Dataset I	Properties
<u>F</u> iles	
Explorer	

8. Select 'Save Display Region to ... ' by clicking on it

Figure 8.36

Options	
O Parameter	ers F1/2 (e.g. used by 'restore display',) [dpl
O Paramete	ers ABSF1/2 (e.g. used by 'absf, apkf')
O Paramete	ers STSR/STSI (used by strip ft)
O Paramete	ers SIGF1,2 (signal region) (used by 'sino')
O Paramete	ers NOISF1,2 (noise region) (used by 'sino')
O A text file	for use with other programs

- 9. Enable 'Parameters F1/2'
- 10. Click on OK
- 11. Type wpar to store the parameter for future use
- 12. Select the user parameter directory

Figure 8.37

Source = C:\Bruker\TopSpin3.0.b.43\exp\stan\nmr\par\user

Figure	8.38
i iguio	0.00

Find file names   enter any string, *, ?     Class =   Dim =   Show Recommended     Type =   SubType =   Reset Filters     1H_256   C13CPD128.mod   C13CPD2K   C13CPD64   C13DEPT135NS32     C13DEPT135p.mod   COSYGP.fixsw   F19_mod   F19COSYGP.test   H1p90_urea     HCCHCOGP3D.pz   HMBCEDETGPI3ND   HSQCEDETGP.mod   HSQCETGPN15   MLEVETGPSW     NP_1H   NP_C13CPD   NP_COSY   NP_DEPTQ   NP_HMBC     NP_HSQC   NP_JRES   NP_ZG30   PRO128PP   PROTON_3exp     proton 90   ROESYETGPSW   SELCOGP.pz   SELNOGP.mod   SELNOGP.mod	The options The	-12		(er (1 op op into: 0.0.40 ic	sip lotari i i in i par laber
Class =   Dim =   Show Recommended     Type =   SubType =   Reset Filters     1H_256   C13CPD128.mod   C13CPD2K   C13CPD64   C13DEPT135NS32     C13DEPT135p.mod   COSYGP.fixsw   F19_mod   F19COSYGP.test   H1p90_urea     HCCHCOGP3D.pz   HMBCEDETGPI3ND   HSQCEDETGP.mod   HSQCETGPN15   MLEVETGPSW     NP_1H   NP_C13CPD   NP_COSY   NP_DEPTQ   NP_HMBC     NP_HSQC   NP_JRES   NP_ZG30   PR0128PP   PROTON_3exp     proton 90   ROESYETGPSW   SELCOGP.pz   SELNOGP.mod	Find file names	enter any string, *, ?			
Type = SubType = Reset Filters     1H_256   C13CPD128.mod   C13CPD2K   C13CPD64   C13DEPT135NS33     C13DEPT135p.mod   COSYGP.fixsw   F19_mod   F19COSYGP.test   H1p90_urea     HCCHCOGP3D.pz   HMBCEDETGPI3ND HSQCEDETGP.mod   HSQCETGPN15   MLEVETGPSW     NP_1H   NP_C13CPD   NP_COSY   NP_DEPTQ   NP_HMBC     NP_HSQC   NP_JRES   NP_ZG30   PRO128PP   PROTON_3exp     proton 90   ROESYETGPSW   SELCOGP.pz   SELNOGP.mod	Class = 🔡 Dim =	Show Recon	nmended		
1H_256     C13CPD128.mod     C13CPD2K     C13CPD64     C13DEPT135NS3       C13DEPT135p.mod     COSYGP.fixsw     F19_mod     F19COSYGP.test     H1p90_urea       HCCHCOGP3D.pz     HMBCEDETGPI3ND     HSQCEDETGP.mod     HSQCETGPN15     MLEVETGPSW       NP_1H     NP_C13CPD     NP_COSY     NP_DEPTQ     NP_HMBC       NP_HSQC     NP_JRES     NP_ZG30     PR0128PP     PROTON_3exp       proton_90     ROESYETGPSW     SELCOGP.pz     SELNOGP.mod	Type = SubTyp	be = 🔽 Reset Fi	Iters		
C13DEPT135p.mod     COSYGP.fixsw     F19_mod     F19COSYGP.test     H1p90_urea       HCCHCOGP3D.pz     HMBCEDETGPI3ND     HSQCEDETGP.mod     HSQCETGPN15     MLEVETGPSW       NP_1H     NP_C13CPD     NP_COSY     NP_DEPTQ     NP_HMBC       NP_HSQC     NP_JRES     NP_ZG30     PR0128PP     PROTON_3exp       proton_90     ROESYETGPSW     SELCOGP.pz     SELNGP.pd     SELNOGP.mod	1H 256	C13CPD128.mod	C13CPD2K	C13CPD64	C13DEPT135NS32
HCCHCOGP3D.pz     HMBCEDETGPI3ND     HSQCEDETGP.mod     HSQCETGPN15     MLEVETGPSW       NP_1H     NP_C13CPD     NP_COSY     NP_DEPTQ     NP_HMBC       NP_HSQC     NP_JRES     NP_ZG30     PRO128PP     PROTON_3exp       proton_90     ROESYETGPSW     SELCOGP.pz     SELNOGP.mod       DET_NO_0_F     SELNOGP.mod     SELNOGP.mod	C13DEPT135p.mod	COSYGP.fixsw	F19_mod	F19COSYGP.test	H1p90_urea
NP_1H     NP_C13CPD     NP_COSY     NP_DEPTQ     NP_HMBC       NP_HSQC     NP_JRES     NP_ZG30     PR0128PP     PROTON_3exp       proton_90     ROESYETGPSW     SELCOGP.pz     SELNOGP.mod       proton_p	HCCHCOGP3D.pz	HMBCEDETGPI3ND	HSQCEDETGP.mod	HSQCETGPN15	MLEVETGPSW
NP_HSQC     NP_JRES     NP_ZG30     PRO128PP     PROTON_3exp       proton_90     ROESYETGPSW     SELCOGP.pz     SELINGP.mod       proton_90     ROESYETGPSW     SELCOGP.pz     SELNOGP.mod	NP_1H	NP_C13CPD	NP_COSY	NP_DEPTQ	NP_HMBC
proton_90 ROESYETGPSW SELCOGP.pz SELMLGP.pz SELNOGP.mod	NP_HSQC	NP_JRES	NP_ZG30	PRO128PP	PROTON_3exp
	proton_90	ROESYETGPSW	SELCOGP.pz	SELMLGP.pz	SELNOGP.mod
SELNOGP.pz SELROGP.pz SI29IGSW SOLVSUP_WET	SELNOGP.pz	SELROGP.pz	SI29IGSW	SOLVSUP_WET	

### 13. Click on Write New...

Figure 8.39



14. Type carbon\_90 in to the new name window



Figure 8.40



16. Select all parameter options

File Options H	olp	Source - C:\Bru	or\TopSpip2.0 b.42\ov	n)stan)nmr\nar\uso
File Options H	eib	Source - C. Ibrui	ker (Topopino.0.b.45/ex	pistaminini panuse
Find file names 🛛 👻	enter any string, *, ?			
Class = 🔽 Dim =	Show Reco	mmended		
Dilli =		innenaea		
Type = 🛛 🝸 SubTy	rpe = 🛛 🖌 Reset f	Filters		
411.050	C12CDD128 mod			
1H_206	C13CPD128.mod	COSVCD firm	C13CPD64	C13DEP1135NS3
H1p00 urea	HCCHCOGP3D pz		HSOCEDETCP mod	HSOCETOPN15
MLEVETGPSW	NP 1H	NP_C13CPD	NP COSY	NP DEPTO
NP HMBC	NP HSOC	NPURES	NP ZG30	PRO128PP
	proton 00	ROESYETGPSW	SELCOGP.pz	SELMLGP.pz
PROTON 3exp	proton 90			

18. Click on Close

# 8.3.4 Determine the 90<sup>0</sup> pulse

1. Click on the 'Aquire' tab in the TopSpin menu bar

Figure 8.42	
<u>Start</u> <u>A</u> cquire <u>P</u> roces	s A <u>n</u> alyse P <u>u</u> blish <u>V</u> iew <u>M</u> anage 🚱
♥ Sample <del>→</del> ₩ <u>L</u> ock V T <u>u</u>	ine 🔻 🕹 Spin 👻 🛱 Shim 👻 🔏 Prosol 💹 Gain 👻 ⊳ Go 👻 Options 👻
2 Click on the down arrow in	side the <b>Co</b>
Figure 8.43	
	Transfer Fid To Disk (tr)
	Estimate Exp. Time (expt)
	Deal Time Co Octure (co)
	Real-Time Go Setup (gs)
	Optimize Acquisition Params (popt)
	Start Automation AU program (xaua)

- 3. Select 'Optimize Acquisition Params (popt)' by clicking on it
- 4. Make the following changes:

OPTIMIZE = Step by step PARAMETER = p1 OPTIMUM = POSMAX STARTVAL = 2 NEXP = 20

VARMOD = LIN	
INC = 2	

Figure 8.44

store as 2D dat	a (ser file)							
The AU program	n specified	I in AUNM will	be executed		WDW= EM			
Perform automa	atic baselin	e correction (	(ABSF)		PH mod= p	ok		
Overwrite existi	na files (di	sable confirma	ation Message)		FT mod= r	10		
Ston sample sn	inning at t	ne end of onti	mization (mash)					
Run optimizatio	n in backg	round	mzation (mash)					
OPTIMIZE	GROUP	PARAMET	OPTIMUM	STARTVAL	ENDVAL	NEXP	VARMOD	INC
Step by step		p1	POSMAX	2		20	LIN	2
Start optimize	Skip curi	ent opti)	Show protocol	Add parame.	Res	store	Save	Read array f

5. Click on Save

NOTE: The ENDVAL parameter has been updated.

6. Click on S	tart optimize
Figure 8.45	
	🔄 poptau 🛛 🔀
	Number of experiments: 20 total experiment time will be: 12 min 0 sec Continue ? [y   n]
	OK Cancel

7. Enter y in to the poptau window



NOTE: The parameter optimization starts. The spectrometer acquires and processes 20 spectra with incrementing the parameter p1 from 2 usec by 2 usec to a final value of 40 usec. For each of the 20 spectra, only the spectral region defined above is plotted, and all the spectra are plotted side-by-side in the file carbon\_90/1/999 as shown in Figure 8.45.



NOTE: The POSMAX value of p1 is displayed in the title which is the 90 degree pulse, along with the experiment number and the NEXP value. Write this value down. To obtain a more accurate 90 degree pulse measurement, follow the steps below.

- 9. Close the popt setup window
- 10. Type re 1 1
- 11. Type p1

12. Enter the value which corresponds to a 360 degree pulse (four times the POSMAX value)

- 13. Type zg
- 14. Type efp

15. Change **p1** slightly and repeat steps 12 and 13, until the signal undergoes a zero crossing as expected for an exact 360 degree pulse.

NOTE: The signal is negative for a pulse angle slightly less then 360 degree and positive when the pulse angle is slightly more then 360 degree.

16. Simply divide the determine 360 degree pulse value by 4. This will be the exact 90 degree pulse length for the proton transmitter on the current probe

### 8.3.5 Observations

# 9 Sensitivity tests

### 9.1 Introduction

This chapter describes the sensitivity test procedures for 1H and 13C. It is assumed that the user is already familiar with acquisition and processing of simple 1D NMR spectra. Chapter 3 (1-D Proton experiment) and chapter 6 (1-D Carbon experiments). Also the 90 degree pulses have to be properly calibrated, Chapter 8 (Determination of the 90 degree pulses)

NOTE: This chapter is intended as a guide for running the 1H and 13C Signal to Noise test on a probe or verifying the values observed using ATP.

### 9.2 1H Sensitivity test

Standard Test Sample:

0.1% Ethylbenzene in CDCl3

### 9.2.1 Experiment setup

1. Click on the 'Start' tab in the TopSpin Menu bar



2. Select Create Dataset by clicking on it

3. Enter the following information in to the 'New' window

	~ ~
Figure	9.2

NAME	proton_se	ensitivity				
EXPNO	1					
PROCNO	1					
DIR	C:\dat	ta3.0		~		
Solvent			CDCI3	~		
Experiment Dirs.	C:\Bruker\	TopSpin3.0.b.4	13\exp\stan\nmr\par	*		
Experiment		PROSE	NS	~		
TITLE						
Proton sensitivit 0.1% Ethylbenze	y test ene in CDCI	3				

NOTE: The directory (DIR) is specific to how the data are stored and therefore may show different entries as the one in Figure 3.4 above. Click on the down arrow button to browse for a specific directory.

- 4. Click on OK
- 5. Click on the 'Aquire' tab in the TopSpin menu bar

Figure 9.3	
<u>Start</u> <u>A</u> cquire <u>P</u> roce	ess A <u>n</u> alyse P <u>u</u> blish <u>V</u> iew <u>M</u> anage 🕜
🕴 Sample 🚽 🗰 Lock 🕅 🤟	T <u>u</u> ne ᢦ 🚯 Sp <u>i</u> n ᢦ 🗣 Shim ᢦ 🔏 Pr <u>o</u> sol 🚾 <u>G</u> ain ᢦ 🕨 Go ᢦ Options ᢦ
6. Select 📲 Sample 🚽 by o	clicking on it
Figure 9.4	
	Turn on sampl <u>e</u> lift air (ej)
	Turn off sample lift air (ij)
	Control sample temperature (edte)

7. Select 'ej' by clicking on it

NOTE: Wait till the sample lift air is turned on and remove any sample which may have been in the magnet.

- 8. Place the sample on too the top of the magnet
- 9. Select 🙀 Sample 🗸 by clicking on it

Figure	9.5
--------	-----



#### 10. Select 'ij' by clicking on it

NOTE: Wait till the sample is lowered down in to the probe and the lift air is turned off. A licking sound may be heard.

<b>[</b>	🔄 Solvents table		×
	△ Solvent	Description	
ľ	Acetic	acetic acid-d4	^
	Acetone	acetone-d6	
	Acetone_Hump	acetone-d6 (CHCl3)	
	C6D6	benzene-d6	
	CD2Cl2	methylenechloride-d2	
	CD3CN	acetonitrile-d3	
	CDCI3	chloroform-d	11
	CDCI3_SENS	chloroform-d (ETB)	
	CH3CN+D2O	HPLC Solvent	
	D20	deuteriumoxide	
	DEE	diethylether-d10	
	Dioxane	dioxane-d8	_
	DME	dimethylether-d6	
	DMF	dimethylformamide-d7	
	DMSO	dimethylsulfoxide-d6	
	EtOD	ethanol-d6	
	H2O+D2O	90%H2O and 10%D2O	~

12. Select 'CDCI3' by clicking on it

13. Select V <u>T</u>une by clicking on it

NOTE: This performs a '**atma**' (automatic tuning) and requires a probe equipped with a automatic tuning module. Other options can be selected by clicking on the down arrow inside the '**Tune**' button.

15. Select	<b>Ֆ</b> Sp <u>i</u> n <del>⊽</del>	by clicking on it
Figure 9.7		
		Turn sample rotation on (ro on)
		Turn sample rotation off (ro off)
		Change sample rotation rate (ro)
		MAS Pneumatic Unit (masdisp)

16. Select 'ro on' by clicking on it

NOTE: Rotation may be turned off for probes such as BBI, TXI, TBI and for small sample probes.

17. Select <mark>국 Shim -</mark> by clicking on it
NOTE: This executes the command ' <b>topshim</b> '. To select other options. click on the down arrow inside the ' <b>Shim</b> ' button.
18. Select <b>Frosol</b> by clicking on it
NOTE: This will load the pulse width and power levels in to the parameter set.

### 9.2.2 Acquisition



NOTE: The relaxation time D1 is by default in this parameter set 60 seconds and therefore the adjustment of the receiver gain will take some time.

2. Select  $\bigcirc$  Go  $\bigtriangledown$  by clicking on it

### 9.2.3 Processing

1. Click on the 'Process' tab in the TopSpin Menu bar

Figure 9.8

<u>S</u> tart	<u>A</u> cquire	Process	A <u>n</u> alyse	P <u>u</u> blish	<u>V</u> iew	<u>M</u> anage	0	
Λ	Spectrum <del>v</del>	Adjust F	Phase 🔻 🛛	Å Calib. A <u>x</u> is <del>–</del>	P	ick P <u>e</u> aks <del>⊸</del>	∫ <u>I</u> ntegrate <del>~</del>	A <u>d</u> vanced <del>▼</del>
_								
- A								

NOTE: This executes a processing program including commands such as an exponential window function 'em', Fourier transformation 'ft', an automatic phase correction 'apk' and a baseline correction 'abs'. Other options are available by clicking on the down arrow inside the 'Proc. Spectrum' button.

### 9.2.4 Calculating the Signal to Noise ratio

The signal to noise ratio is determined on the intensity of the quartet lines between 2ppm and 3ppm. It is calculated by AU-program **sinocal** over a range of 2ppm between

2.8ppm and 7ppm. The s/n ratio is strongly dependant on good resolution and lineshape. The splitting between the two central lines of the methylquartet should go lower than 15% (with LB=1Hz), see Figure 9.9.



#### 1. Type sinocal on the command line

Figure 9.10

😂 sinocal	
Enter left limit of signa	al range in ppm :
3	
	OK Cancel

2. Enter 3 for the left limit of the signal range

3. Click on OK	
Figure 9.11	
	🔄 sinocal 🛛 🔀
	Enter right limit of signal range in ppm :
	OK Cancel

4. Enter 2 for the right limit of the signal range

5. Click on OK	
Figure 9.12	
	🥌 sinocal 🛛 🔀
	Enter left limit of noise range in ppm :
	7
	OK Cancel

6. Enter 7 for the left limit of the noise range

7. Click on OK	
Figure 9.13	
	🔄 sinocal 🛛 🔀
	Enter right limit of noise range in ppm :
	OK Cancel

8. Enter 2.8 for the right limit of the noise range

9. Click on OK	]	
Figure 9.14		
	💐 sinocal	
	Enter noise width in ppm :	
	2	
		OK Cancel

- 10. Enter 2 for the noise width
- 11. Click on OK



### 9.2.5 Observations

# 9.3 13C Sensitivity test with 1H decoupling

Standard Test Sample:

10% Ethylbenzene in CDCl3

### 9.3.1 Experiment setup

1. Click on the 'Start' tab in the TopSpin Menu bar

Figure 9.16					
<u>S</u> tart <u>A</u> c	quire <u>P</u> rocess	A <u>n</u> alyse F	P <u>u</u> blish <u>V</u> ie	ew <u>M</u> anage	2
	Create Dataset	Find Dataset	🎯 Open <u>D</u> a	ataset 📘 Past	te Dataset 🔡 Read Pars.
2. Select	eate Dataset	by clicking	on it		
3. Enter the follow	ing informatio	n in to the 'N	New' windo	ow	
-	-				
Figure 9.17					
	🔤 New				
	Prepare for a new	vexperiment by cre	eating a new da	ita set and	
	For multi-receiver	experiments accor	rding to the sele ral datasets are	ected experiment t	ype.
	Please define the	number of receive	ers in the box be	elow.	
	NAME	carbon_sens_e	tb		
	EXPNO	1			
	PROCNO	1			
	DIR	C:\data3.0			*
	Solvent	- 12		CDCI3	*
	Experiment Dirs.	C:\Bruker\TopSp	pin3.0.b.43\exp\	\stan\nmr\par	*
	Experiment		C13SENS		*
	TITLE				
	13C sensitivity t	est with 1H decoup	oling		
	10% Ethylbenze	ene in CDCI3			
	Show new da	taset in new windo	W		
		(1.0. 16)			
	1 Receivers	(1,2, 16)			
		ОК	Cancel Mo	ore Info He	lp

NOTE: The directory (DIR) is specific to how the data are stored and therefore may show different entries as the one in Figure 3.4 above. Click on the down arrow button to browse for a specific directory.

- 4. Click on OK
- 5. Click on the 'Aquire' tab in the TopSpin menu bar

#### Figure 9.18

	-											
		<u>S</u> tart	<u>A</u> cquire	Process	A <u>n</u> alyse	P <u>u</u> blish	<u>V</u> iew	<u>M</u> anage	2			
	ţ	Sample	e <del>▼</del>   ∰ Loo	k V T <u>u</u> ne ₹	Spin	▼ 🛱 Shim	-	Pr <u>o</u> sol	<u>G</u> ain <del>▼</del>	▶ Go 🗢	Options 🗢	

6. Select <table-of-contents> Sample 😽</table-of-contents>	by clicking on it
Figure 9.19	
	Turn on sampl <u>e</u> lift air (ej)
	Turn off sample lift air (ij)
	Control sample temperature (edte)

7. Select 'ej' by clicking on it

NOTE: Wait till the sample lift air is turned on and remove any sample which may have been in the magnet.

8. Place the sample on too the top of the magnet



10. Select 'ij' by clicking on it

NOTE: Wait till the sample is lowered down in to the probe and the lift air is turned off. A licking sound may be heard.

11. Select	E Lock	by clicking on it
------------	--------	-------------------

Figure 9.21

🥌 Solvents table		×		
△ Solvent	Description			
Acetic	acetic acid-d4	^		
Acetone	acetone-d6			
Acetone_Hump	acetone-d6 (CHCI3)			
C6D6	benzene-d6			
CD2Cl2	methylenechloride-d2			
CD3CN	acetonitrile-d3			
CDCI3	chloroform-d			
CDCI3_SENS	chloroform-d (ETB)			
CH3CN+D2O	HPLC Solvent			
D2O	deuteriumoxide			
DEE	diethylether-d10			
Dioxane	dioxane-d8			
DME	dimethylether-d6			
DMF	dimethylformamide-d7			
DMSO	dimethylsulfoxide-d6			
EtOD	ethanol-d6	12		
H2O+D2O	90%H2O and 10%D2O	~		
Lock nucleus: 2H	Canc	el		

12. Select 'CDCI3' by clicking on it

13. Select V Tune → by clicking on it

NOTE: This performs a '**atma**' (automatic tuning) and requires a probe equipped with a automatic tuning module. Other options can be selected by clicking on the down arrow inside the '**Tune**' button.

15. Select	& Sp <u>i</u> n <del>▼</del>	by clicking on it
Figure 9.22		
		Turn sample rotation on (ro on)
		Turn sample rotation off (ro off)
		Change sample rotation rate (ro)
		MAS Pneumatic Unit (masdisp)

16. Select 'ro on' by clicking on it

NOTE: Rotation may be turned off for probes such as BBI, TXI, TBI and for small sample probes.

17. Select	Shim → by clicking on it
NOTE: This arrow inside	s executes the command ' <b>topshim</b> '.To select other options. click on the dowr e the ' <b>Shim</b> ' button.
18. Select	feresol by clicking on it

NOTE: This will load the pulse width and power levels in to the parameter set.

### 9.3.2 Acquisition

1. Select Gain - by clicking on it

NOTE: The relaxation time D1 is by default in this parameter set 300 seconds and therefore the adjustment of the receiver gain will take some time.



### 9.3.3 Processing

1. Click on the 'Process' tab in the TopSpin Menu bar

Fig	ure 9.2	23								
		<u>S</u> tart	<u>A</u> cquire	Process	A <u>n</u> alyse	P <u>u</u> blish	<u>V</u> iew	<u>M</u> anage	0	
		Λ Pro <u>c</u>	. Spectrum <del>v</del>	Adjust F	Phase 🗢 🎤	<mark>∖</mark> Calib. A <u>x</u> is <del>⊲</del>	Pi	ick P <u>e</u> aks <del>→</del>	∫ <u>I</u> ntegrate <del>~</del>	A <u>d</u> vanced <del>▼</del>
2.	Click	on	A Pro <u>c</u> .	Spectru	m 👻					

NOTE: This executes a processing program including commands such as an exponential window function 'em', Fourier transformation 'ft', an automatic phase correction 'apk' and a baseline correction 'abs'. Other options are available by clicking on the down arrow inside the 'Proc. Spectrum' button.

### 9.3.4 Calculating the Signal to Noise ratio

The signal to noise ratio is determined on the highest peak of the aromatic part between 127ppm and 129ppm see Figure 9.24. below. It is calculated by AU-program **sinocal** over a range of 40ppm between 30ppm and 125ppm. The s/n ratio is strongly dependent on good resolution and line shape.



#### 1. Type sinocal on the command line

💐 sinocal 🛛 🔀
Enter left limit of signal range in ppm :

2. Enter 128 for the left limit of the signal range

3. Click on o	ĸ
Figure 9.26	
	🔄 sinocal 🛛 🔀
	Enter right limit of signal range in ppm :
	127
	OK Cancel

4. Enter 127 for the right limit of the signal range

5. Click on OK	]
Figure 9.27	
	🔄 sinocal 🛛 🔀
	Enter left limit of noise range in ppm :
	125
	OK Cancel

6. Enter 125 for the left limit of the noise range

7. Click on OK	Ĵ
Figure 9.28	
	🤹 sinocal 🛛 🔀
	Enter right limit of noise range in ppm : 30 OK Cancel

- 8. Enter 30 for the right limit of the noise range
- 9. Click on OK

Figure	9.29
--------	------

🖕 sinocal	X
Enter noise width in ppm :	
40	
	OK Cancel

10. Enter 40 for the noise width

No acquisition r	unning: C:/data/pz/nm <u>r/carbo</u>	n_sens/1/pdata/1			
pectrum Proc	Pars AcquPars Title Pu	ulseProg Peaks Integrals Sa	Imple Structure Fid Acqu		
[		1			
<u>.</u>					
-					
_					
-					
-					
		inocal	xt sino value: 117.6		
		einocal	est sino value: 117.6 gnal from 129.00 to 127.00 ppm lise from 125.00 to 85.00 ppm	<b>3</b>	
		Be Sh No	est sino value: 117.6 gnal from 129.00 to 127.00 ppm lise from 125.00 to 85.00 ppm Close Details	O O	
2 =		e sinocal	est sino value: 117.6 gnal from 129.00 to 127.00 ppm lise from 125.00 to 85.00 ppm		
			est sino value: 117.6 gnal from 129.00 to 127.00 ppm lise from 125.00 to 65.00 ppm Close Details		
			est sino value: 117.6 gnal from 129.00 to 127.00 ppm lise from 126.00 to 85.00 ppm		
			est sino value: 117.6 gnal from 129.00 to 127.00 ppm lise from 125.00 to 85.00 ppm		
			est sino value: 117.6 gnal from 129.00 to 127.00 ppm lise from 125.00 to 85.00 ppm Close Details		

### 9.3.5 Observations

# 9.4 13C Sensitivity test without 1H decoupling

Standard Test Sample:

ASTM (60% C6D6 / 40% p-Dioxane)

### 9.4.1 Experiment setup

1. Click on the 'Start' tab in the TopSpin Menu bar

Figure 9.31	e <u>P</u> rocess ate Dataset	A <u>n</u> alyse F <u>i</u> nd Datase	P <u>u</u> blish <u>v</u> t 🏹 Open [	<u>/</u> iew <u>)</u> ataset	Manage	2 Dataset 📄 Read Pars.
2. Select Create	e Dataset	by clicking	g on it			
3. Enter the following	information	in to the	'New' wind	wob		
<b>F</b> i 0.00						
Figure 9.32	-				-	<b>7</b> 1
P ir F P	Prepare for a new on nitializing its NMR p For multi-receiver of Please define the r	experiment by coarameters acc experiments sev number of recei	creating a new of ording to the serveral datasets a vers in the box	lata set a elected e re create below.	and xperiment typ ed.	e.
	FXPNO	carbon_sens	_asun			-
	PROCNO	1				-
	DIR	C:\data3.0	)			~
	Solvent	e e e e e e e e e e e e e e e e e e e		CDC	:13	~
1	Experiment Dirs.	C:\Bruker\Top	Spin3.0.b.43\ex	p\stan\n	mr\par	<b>~</b>
	Experiment TITLE		C13SENS			<b>~</b>
	13C sensitivity te ASTM (60% C6D	st no 1H decou 6 / 40% Dioxan	pling e)			
	Show new data	aset in new win 1,2,16)	dow			
		ОК	Cancel	More Info	b Help	

NOTE: The directory (DIR) is specific to how the data are stored and therefore may show different entries as the one in Figure 3.4 above. Click on the down arrow button to browse for a specific directory.

4. Click on OK

5. Click on the 'Aquire' tab in the TopSpin menu bar

Figure 9.33

۰.									
	<u>S</u> tart	Acquire	Process	A <u>n</u> alyse I	P <u>u</u> blish \	<u>/</u> iew	<u>M</u> anage	2	
	🕴 Sampl	e 🚽 🌞 Loo	k V T <u>u</u> ne	▼ 🕹 Sp <u>i</u> n マ	Shim -	₽ ft P	r <u>o</u> sol 🚾	<u>G</u> ain <del>▼</del>	► Go 🗢 Options 🗢

6. Select	📑 Sample 😽	by clicking on it
Figure 9.34		
		Turn on sampl <u>e</u> lift air (ej)
		Turn off sample l <u>i</u> ft air (ij)
		Control sample temperature (edte)

7. Select 'ej' by clicking on it

NOTE: Wait till the sample lift air is turned on and remove any sample which may have been in the magnet.

8. Place the sample on too the top of the magnet



10. Select 'ij' by clicking on it

NOTE: Wait till the sample is lowered down in to the probe and the lift air is turned off. A licking sound may be heard.

5	-		-
	Solvents table		×
	△ Solvent	Description	
	Acetic	acetic acid-d4	^
	Acetone	acetone-d6	
	Acetone_Hump	acetone-d6 (CHCl3)	
	CGDG	benzene-d6	
	CD2Cl2	methylenechloride-d2	
	CD3CN	acetonitrile-d3	
	CDCI3	chloroform-d	
	CDCI3_SENS	chloroform-d (ETB)	
	CH3CN+D2O	HPLC Solvent	
	D20	deuteriumoxide	
	DEE	diethylether-d10	
	Dioxane	dioxane-d8	
	DME	dimethylether-d6	
	DMF	dimethylformamide-d7	
	DMSO	dimethylsulfoxide-d6	
	EtOD	ethanol-d6	
	H2O+D2O	90%H2O and 10%D2O	

12. Select 'C6D6' by clicking on it

13. Select by clicking on it ν Tune 🗢

NOTE: This performs a 'atma' (automatic tuning) and requires a probe equipped with a automatic tuning module. Other options can be selected by clicking on the down arrow inside the 'Tune' button.

15. Select by clicking on it 🕹 Spin 🗸 Turn sample rotation on (ro on) Turn sample rotation off (ro off) Change sample rotation rate (ro) MAS Pneumatic Unit (masdisp)

16. Select 'ro on' by clicking on it

Figure 9.37

NOTE: Rotation may be turned off for probes such as BBI, TXI, TBI and for small sample probes.

17. Select 독 Shim ▼ by clicking on it

NOTE: This executes the command 'topshim'. To select other options. click on the down arrow inside the 'Shim' button.



NOTE: This will load the pulse width and power levels in to the parameter set.

- 19. Select the 'AcquPars' tab by clicking on it
- 20. Make the following changes:

PULPROG = zgTD = 64kSW [ppm] = 200 O1p = 100

- 21. Select the 'ProcPars' tab by clicking on it
- 22. Make the following changes:

SI = 32k

LB [Hz] = 3.5

23. Click on the 'Aquire' tab in the TopSpin menu bar

### 9.4.2 Acquisition

1. Select	<u> </u>	by clicking on it
NOTE: The fore the a	ne relaxation ti djustment of th	me D1 is by default in this parameter set 300 seconds and there- ne receiver gain will take some time.
2. Select	D Go 🗸	by clicking on it

### 9.4.3 Processing

1. Click on the 'Process' tab in the TopSpin Menu bar

Fig	Figure 9.38									
		<u>S</u> tart	<u>A</u> cquire	Process	A <u>n</u> alyse	P <u>u</u> blish	<u>V</u> iew	<u>M</u> anage	0	
		A Pro <u>c</u>	Spectrum <del>v</del>	Adjust P	hase 🗢 🏼 🍌	Calib. A <u>x</u> is <del>⊲</del>	Pi 🎊 Pi	ck P <u>e</u> aks <del>⇒</del>	∫ <u>I</u> ntegrate <del>→</del>	A <u>d</u> vanced <del>▼</del>
2.	Click	on 🧧	A Pro <u>c</u> .	Spectru	m 👻					

NOTE: This executes a processing program including commands such as an exponential window function 'em', Fourier transformation 'ft', an automatic phase correction 'apk' and a baseline correction 'abs'. Other options are available by clicking on the down arrow inside the 'Proc. Spectrum' button.

### 9.4.4 Calculating the Signal to Noise ratio

The signal to noise ratio is determined on the triplet of the deuterated benzene between 127ppm and 129ppm. It is calculated by AU-program **sinocal** over a range of 40ppm between 70ppm and 125ppm. The s/n ratio is strongly dependent on good resolution and line shape. The splitting of the 1:1:1 triplet should go lower than 9% (5mm) see Figure 9.18. 10% (10mm) and 12% (20mm).



### 1. Type sinocal on the command line

#### Figure 9.40

×
ange in ppm :
OK

2. Enter 128 for the left limit of the signal range

3. Click on OK	
Figure 9.41	
	🔄 sinocal 🛛 🔀
	Enter right limit of signal range in ppm :      127
	OK

- 4. Enter 127 for the right limit of the signal range
- 5. Click on OK

Figure 9.42	💩 sinocal 🛛 🗙
	Enter left limit of noise range in ppm :
	125
	OK Cancel

6. Enter 125 for the left limit of the noise range

7. Click on Or	
Figure 9.43	
	🔄 sinocal 🛛 🔀
	Enter right limit of noise range in ppm :
	70
	OK Cancel

8. Enter 30 for the right limit of the noise range

9. Click on OK	]	
Figure 9.44		
	💐 sinocal	
	Enter noise width in ppm :	
		OK Cancel

- 10. Enter 40 for the noise width
- 11. Click on OK



### 9.4.5 Observations

#### B7169\_00\_01
# **10** Spectrometer configuration

# 10.1 Hardware Configuration

Fig	ure 10.1									
		<u>S</u> tart	<u>A</u> cquire	<u>P</u> rocess	A <u>n</u> aly	se P <u>i</u>	ublish	<u>V</u> iew	Manage	0
			Pr <u>e</u> ference	s Spectr <u>o</u>	ometer <del>–</del>	Security	<u>/</u>	ommands 🗟	Remote	
2. Fia	Click on ure 10.2	the dov	wn arrow in	side the	Spectr <u>o</u>	meter <del>v</del>	butto	n		
0		Hard	ware Detection	Þ	Configure	Hardware	(cf)			
		Expe	riments/Param	eters 👂	Initialize S	pectromet	er Interfa	ce (ii)		
		Save	/Restore Instal	lation 👂	Edit the P	robehead	Table (ec	dhead)		
		Spec	trometer Usag	e (account)	Setup Lin	earization	Correctio	n Tables (co	ortab)	
					Find Ethe	rnet Addre	sses (ha	)		

1. Click on the 'Manage' tab in the TopSpin Menu bar

3. Select 'Hardware Detection'

- 4. Select 'Configure Hardware (cf)' by clicking on it
- 5. Enter the NMR administration password

6. Click on OK

opoon on otor configuration (of			
	ų.		
Configure the hardware of you	r spectrometer or create a co	onfiguration for a datasta	ation.
Active configuration: "spect"			
,			
Available spectrometer confid	urations		
	Jananono		1000000
A Configuration	Spectrometer type	Frequency [MHZ]	Туре
Bruker_default_av500	Avance-AV 500	500.13	Datastation
Bruker_default_avii/00	Avance III 600	700.13	Datastation
Druker_deladit_aviilooo	Avalice il 000	000.13	Datastation
Select one of the available con	figurations or create a new s	pectrometer configuration	on!
Select one of the available con	figurations or create a new s	pectrometer configuratio	on!
Select one of the available con Press "Edit" to modify or use a	figurations or create a new s existing configuration (e.g.	pectrometer configuratio	on! nardware
Select one of the available con Press "Edit" to modify or use a or if you want to use the config Tress "New" to create a new st	figurations or create a new s n existing configuration (e.g. uration from a previous Top5	pectrometer configuratio if you want to add new h pin version).	on! hardware
Select one of the available con Press "Edit" to modify or use a or if you want to use the config Press "Neiver" to detee the se	figurations or create a new s n existing configuration (e.g. uration from a previous TopS ectrometer configuration fro lected configuration.	pectrometer configuratio If you want to add new h pin version). m scratch.	on! hardware
Select one of the available con Press "Edit" to modify or use a or if you want to use the config Press "New" to create a new sp Press "Show" to delete the se Press "Show" to were the "Conf	figurations or create a new s n existing configuration (e.g. uration from a previous Tops bectrometer configuration. liguration summary' (uxmm: ir	pectrometer configuratio if you want to add new h spin version). m scratch. ifo) of the selected conf	on! nardware iguration.

- 4. Select Configuration for 'Spect' by clicking on it
- 5. Click on Edit

Figure	10.4	

e	Cf		×
	Edit configuration:		
	Configuration Configuration name Spectrometer Datastation 1H frequency of magnet [MHz]	spect © 0 300.13	
	Debug Use debug module		
-			<pre></pre>
L .			
6. Click on Next >	]		

Figure 10.5

HDDD Dreamplifier	thv10	~			
BSMS Smart Magnet Control System	140 236 00 20				
VTI I Variable Temperature Unit	143.200.33.20				
CNMP Software HyStar	100	-			
Gradient Temperature Unit (BCU-20)	00	~			
MAS Pneumatic Control Unit	no	~			
Bruker Automatic Changer	ttv04	~			
Barcode Printer	00	~			
Crvo Controller	no	~			
HPCU High Power Control Unit	no	~			
Preemphasis/Gradient Unit	no	~			
Fast Gradient Supervisor	no	~			
Gradient Power Supply Control Unit	no	~			
Radio Frequency Supervisor	no	~			
Laskouitek					
LOCKSWIICH		Tana			
2H Lockswitch connected to Amplifier	at Blanking Signal	no	~		
19F Lockswitch connected to Amplifier	r at Blanking Signa	I no	~		

7. Enter the RS232 ports for the external devices as shown in Figure 10.5

NOTE: Use the default connection listed on the label on the inside of the console. If a BACS 60 or 120 is used, select the proper RS232 port (normally tty08) and be sure the power of the BACS is on. The sample changer will configure the correct number of sample holders. If a SIXPACK, CASE, MAS or HRMAS sample changer is used, set the BACS port to an unused port number (for example tty20. After a few seconds a message will appear that there is no communication to the sample changer and a default of 60 sample holders is been used. Just click on the 'OK' button to continue with cf. The number of sample holders for different sample changers can be set in the ICONNMR configuration (Default Number of Sample Holders)

8. Click on Next >

Additional configuration:	
Security configuration	
Enable peak power check (POWCHK):	
Sample changer configuration	
Should the sample changer control the lift:	
Delay between SX and next command [s]:	20
BACS, SampleRail or SampleJet options:	
Fast SampleRail mode:	

NOTE: If the Power check and Cortab have been performed on the system, enable the peak power check (POWCHK). If the Power check has not been performed do not use this option.

9. Click on Figure 10.7

Next >

△ Nucleu	s Name	Receptivity (rel. 13C)	Spin	Frequency (rel. 1H)	
1H	Hydrogen	5680.0	1/2	300.13	1
2H	Deuterium	0.00821	1	46.071782	
зн	Tritium	1.0E-9	1/2	320.130585	
3He	Helium	0.00326	1/2	228.637344	
6Li	Lithium	3.58	1	44.167389	
7LI	Lithium	1540.0	3/2	116.641914	
9Be	Beryllium	78.8	3/2	42.173706	
10B	Boron	22.1	3	32.244941	
11B	Boron	754.0	3/2	96.293631	
13C	Carbon	1.0	1/2	75.467749	
14N	Nitrogen	5.69	1	21.681046	
15N	Nitrogen	0.0219	1/2	30.411909	
170	Oxygen	0.0611	5/2	40.686994	
19F	Fluorine	4730.0	1/2	282.404355	
21Ne	Neon	0.0359	3/2	23.693151	
23Na	Sodium	525.0	3/2	79.390087	
25Mg	Magnesium	1.54	5/2	18.372863	
27AI	Aluminum	1170.0	5/2	78.204451	
2951	Silicon	2.09	1/2	59.627388	
31P	Phosphorus	377.0	1/2	121.494851	
33S	Sulfur	0.0973	3/2	23.037979	
35CI	Chlorine	20.2	3/2	29.406464	
37CI	Chlorine	3.77	3/2	24.477777	
39K	Potassium	2.69	3/2	14.005185	
41K	Potassium	0.0328	3/2	7.687245	
43Ca	Calcium	0.0527	7/2	20.198836	

10. Click on Restore 11. Click on Save 12. Click on Next >	
Figure 10.8	
frequency log	jcal amplifier preamplifier
BF1 0.0 MHz NU SF01 0.0 MHz IU OFS1 0.0 HHz BF2 0.0 MHz NU SF02 0.0 MHz IU OFS2 0.0 MHz IV	C1 F1 SGU1 X 150 W 1H C2 F2 SGU2 2H 100 W
	settings

NOTE: The edsp window should show the connections from the Amplifiers to the Preamplifiers only. If there are incorrect connections, click on 'CLEAR PREAMPLIFIER CONNECTIONS' and draw the correct connections.

13. Click on Save	]
Figure 10.9	
-	or 🛛
-	
	Configuration summary (uxnmr.info).
	CONFIGURATION INFORMATION
	Parts
	Date : Thu Jul 22 13:56:07 2010
	Release : TopSpin Acquisition Version ts_3_0-pl2
	Installed in : C:/Bruker/TopSpin3.0.b.43
	Host : 561ADFB7F87F45B
	OS : Windows XP (Vs 5.1) Service Pack 3
	CPU : Intel(R) Core(TM)2 Duo CPU E4500 @ 2.20GHz (2 cores at 2200 MHz
	User : pz
	1H-frequency : 300.13 MHz
	Configured in: C:/Bruker/TopSpin3.0.b.43/conf/instr/spect
	IPSO: connected to spectrometer subnet
	- TCP/IP address = 149.236.99.254
	- Tetrl : 1
	- Fctrls: 2
	- Gctrll: without digital preemphasis
	- RCTI : I
	DRU: AOS DRU Z100977/00767 ECL 04.00
	- TCP/IP address = 149.236.99.89
	- Firmware Version = 100209
	- DRU controls AQS-Rack and HPPR/2
	AQS: connected to 149.236.99.89:/dev/tty10
	_Slot_SBSBBoard
	Number Addr Type HW-VS FW-VS ID ECL Name Description
	2 0x10 0x72 0x4 AU R 4.2 REC-1 AQS RXAD400 Z102116/1002 ECL 04.
	3 Ox34 Ox2 Ox1 X 5.0 REF-1 REF-400 Reference Board for AQS 🗸
	8
	< Previous Next > Cancel Print

the configuration information with the installation data
14. Click on Print 15. Click on Next >
Figure 10.10
S S S
Additional configuration programs:
Config Installation of standard experiments Expinstall Solvent table setup Edsolv Probe table setup Edhead Lock parameter setup Edickk Spectrometer parameters setup Edscon

NOTE: The configuration information is displayed on the screen. Store the print out of

# 10.2 Expinstall

- 1. Click on Expinstall
- 2. Enter the NMR administration password
- 3. Click on OK

Figure	10	.1	1

		Expinstall		
Expinstall in: parameter s or datastatic installation of For a custor spectromete to <topspin< td=""><td>stalls pulse p ets and vari- on usage. It i of TOPSPIN. nized datast r configurati installation</td><td>orograms, A ous other re must be per For spectro ation config ion directory dir.&gt;/conf/in</td><td>U programs, sources for s formed once a meter control uration copy y ( (typically cal str.</td><td>pectromete after the do cf first. your led "spect")</td></topspin<>	stalls pulse p ets and vari- on usage. It i of TOPSPIN. nized datast r configurati installation	orograms, A ous other re must be per For spectro ation config ion directory dir.>/conf/in	U programs, sources for s formed once a meter control uration copy y ( (typically cal str.	pectromete after the do cf first. your led "spect")
WARNING: Please arch AU-program "expinstall".	ve all your № s and PULS	NODIFIED B E-programs	ruker PARAM before runnir	IETER-files, 1g
	< Back	Next >	Finish	Cancel

### 4. Click on Next >

Figure 10.12

Experiment installation and AU compilation	X
Select the type of installation:	
<ul> <li>Installation for Datastation (Default)</li> </ul>	
<ul> <li>Installation for Datastation (Customize)</li> </ul>	
<ul> <li>Installation for Spectrometer</li> </ul>	
< Back Next > Finish Cano	el

5. Select 'Installation for Spectrometer'

Figure 10.13	Expinstall for Spectrometer Select the type of acquisition: U High Resolution Systems Solid State Systems
	Select the type of acquisition: Image: High Resolution Systems Image: Solid State Systems
	Select the type of acquisition: I High Resolution Systems Solid State Systems
	Solid State Systems
	Micro-Imaging and Diffusion Systems

7. Select 'High Resolution System'

2

8. Click on Next >

🖨 Expinstall for Spectrometer 🛛	×
Select the items you want to install:	
Install Pulse Programs	
Install Bruker AU Programs	
Recompile All User AU Programs	
Install Library CPD Programs	
Install Library Gradient Files	
Install Library Shape Files	
Convert Standard Parameter Sets	
Install Standard Scaling Region Files	
Install Bruker Macros	
Install Bruker Python Programs	
Select all Select none	
< Back Next > Finish Cancel	

11. Click on Next >	•
Figure 10.15	
	Expinstall for Spectrometer
	Select your printer:
	Default printer. HP LaserJet 2100 PCL6
	Select your plotter:
	Default plotter: HP LaserJet 2100 PCL6
	Select the plotter paper format:
	Paper format: A4 / Letter
	< Back Next > Finish Cancel

12. Select Default printer and plotter

13. Select Paper format

14. Click on Next >

Figure	10.1	16
		•••

Select the basic frequency	of your spectrometer:
Basic frequency (MHz):	300.13
Select the digitizer:	
Type of digitzer:	DRU 👱
Select the acquisition mode	6
Acquisition mode:	DQD 🗸
Select the pre-scan-delay [	DE:
Default pre-scan-delay (µ	s): 6.5

16. Click on Next >

Figure 10.17

Expiriation will be exect	aced with following options.
Installation for Spe	ctrometer
(High Resolution)	
Configuration name	e: spect
Install Pulse Progra	ams
Install Bruker AU P	rograms
Install Library CPD	Programs
Install Library Grad	lient Files
Install Library Shap	be Files
Convert Standard I	Parameter Sets
Install Standard Sc	aling Region Files
Install Bruker Macr	os
Install Bruker Pytho	on Programs
Basic frequency:	300.13 MHz
Digitizer:	DRU
Acquisition mode:	DQD
Pre-scan-delay:	6.5 µs
Printer:	HP LaserJet 2100 PCL6
	HP LaserJet 2100 PCL6
Plotter:	

17. Click on Finish

NOTE: expinstall starts now. This process will take approximately. 2 Minutes. On finish the message below appears (Figure 10.17.). To set up a time schedule to perform an NMR\_save periodically (recommended) follow the instructions in 10.3 Set up the cron job for NMR\_save.

🔄 Cron check 🛛 🔀
An automatic periodical backup of your TopSpin configuration can be defined in TopSpin. Currently you do not use this tool. Press "Automatic Backup" to open the configuration tool.
□ Do not show this message again
Help Automatic Backup Close

## **10.3** Set up the cron job for NMR\_save

1. Click on Automatic Backup	
Figure 10.19	
NMR_Save	
Save installation files Restore installat	tion files Save user files Restore user files
Save installation specific files. Installation specific files are collected a to copy the files from a previous install of the installation specific files. Note: To save user specific files use the "Sa	and stored into a compressed file. This compressed file can be used lation to a new installation or to create a backup we user files" tab.
Location of backup file: Overwrite existing backup file: Installation to be saved (TopSpin hom Spectrometer configuration (e.g. spec Display default information: Display additional information:	C:\Bruker\TopSpin3.0.b.43\nmr_backup  te): C:\Bruker\TopSpin3.0.b.43  tp: spect
Execute "Save installation specific file	s" periodically Automatic Backup
	Save Close

2. Click on Automatic Ba	ackup
Figure 10.20	
New periodical	
Job Command	.nmrsave -date -path "C:\Bruker\TopSpin3.0.b.43\nmr_backup" -source "C:\
Execution scope	TopSpin (requires authentication)
Options          Image: Options <t< td=""><th>execution on</th></t<>	execution on
Minute of the I	hour 🕑 from: 12 💌 to: Ignore 👻 +
Hour of the da	y y from: 14 v to: Ignore v + -
Day of the mo	nth v from: 22 v to: Ignore v + -
Month of the y	ear 🕑 from: * 💌 to: Ignore 🖌 + -
Day of the wee	ek 🔹 from: * 💌 to: Ignore 👻 + - 👻
	OK Cancel

NOTE: In this example an NMR\_save is performed from January to December on the 1st day of the month at 2 o'clock in the morning.

3. Click on OK

Figure 10.21		
	🤤 Cf	X
	Additional configuration programs:	
	Config	
	Installation of standard experiments	Expinstali
	Solvent table setup	Edsolv
	Probe table setup	Edhead
	Lock parameter setup	Edlock
	Spectrometer parameters setup	Edscon
		< Previous Finish > Cancel

# **10.4** Selection of current Probehead

### **10.4.1** Current probe equipped with pics:

1. Click on Ec	lhead	
Figure 10.22		
	🔌 edhead	
	<u>D</u> ptions	<u>E</u> xit
	Current probe: 5 mm PABBO BB-1H/D Z-GRD Z104275/0007	[36]
	8 mm 1H	[02]
	5 mm Dual 13C/1H	[03]
	5 mm Dual 19F/1H	[04]
	5 mm QNP 1H/15N/13C/31P Z3246/0046	[05]
	5 mm QNP 1H/13C/31P/19F	[06]
	5 mm QNP 1H/13C/31P/19F Z-grad	[07]
	5 mm QNP 1H/29Si/13C/31P Z00010/1	[08]
	5 mm Multinuclear Z0000/1	[09]
	5 mm Multinuclear inverse	[10]
	5 mm Multinuclear inverse Z-grad	[11]
	5 mm PABB0 BB-1H/D Z-GRD Z104275/0007	[36] 😈
	Define as current probe Edit Probe Parameters	Exit

NOTE: The new probe is been automatically added to the probehead list.

2.Click on Define as current probe

Figure 10.23	
	current probe
	5 mm PABBO BB-1H/D Z-GRD Z104275/0007
	seen
3. Click on <b>seen</b>	
4. Click on Exit	
Figure 10.24	
🧔 Edit S	pestrometer Parameter X
	1H/2H 1H BB 1 Inner Coll
	1H/2H 2H     1H     1 Outer Coll       XBB19F 2HS     D     2 Outer Coll
	Save Clear cable connections Info Param Close
NOTE: If desired, the	connections of the preamplifiers to the probe can be changed

10.4.2 Current probe not equipped with pics and with probe parameters:

1. Type <mark>edhead</mark>		
Figure 10.25		
	🧶 edhead	
	Options	<u>E</u> xit
	Current probe: 5 mm Dual 13C/1H Z000001/0001 [03]	
	8 mm 1H	[02]
	5 mm Dual 13C/1H Z000001/0001	[03]
	5 mm Dual 19F/1H	[04]
	5 mm QNP 1H/15N/13C/31P Z3246/0046	[05]
	5 mm QNP 1H/13C/31P/19F	[06]
	5 mm QNP 1H/13C/31P/19F Z-grad	[07]
	5 mm QNP 1H/29Si/13C/31P Z00010/1	[08]
	5 mm Multinuclear Z0000/1	[09]
	5 mm Multinuclear inverse	[10]
	5 mm Multinuclear inverse Z-grad	[11]
	5 mm PABBO BB-1H/D Z-GRD Z104275/0007	[36] 🥃
	Define as current probe Edit Probe Parameters	Exit

- 2. Select current probehead from the list by clicking on it
- 3. Click on Define as current probe

5. Click on Save

Figure 10.26			
	🍓 current pro	be (	
	្ដ្ឋី Current រី 5 mm Di	Probe is no <del>w</del> : ual 13C/1H Z0000	00170001
		seen	
4. Click on seen	]		
5. Click on Exit	]		
Figure 10.27			
🧔 Edit Spectron	neter Parameter		X
F	Preamplifier	Probe	
	1H/2H 1H 1H/2H 2H XBB19F 2HS	13C 1 Inner 1H 1 Oute 2H 2 Oute	r Coll
		Save Clear cable connect	ions Info Param Close

NOTE: If desired, the connections of the preamplifiers to the probe can be changed

5. Click on Save

# **10.4.3** Current probe not equipped with pics and without probe parameters:

#### 1. Type edhead

Iptions	<u>E</u> >
Current probe: 5 mm Dual 19F/1H [04]	
8 mm 1H	[02
5 mm Dual 13C/1H 2000001/0001	[03
5 mm Dual 19F/1H	[04
5 mm QNP 1H/15N/13C/31P Z3246/0046	[05
5 mm QNP 1H/13C/31P/19F	[06
5 mm QNP 1H/13C/31P/19F Z-grad	[07
5 mm QNP 1H/29Si/13C/31P Z00010/1	[08
5 mm Multinuclear Z0000/1	[09
5 mm Multinuclear inverse	[10
5 mm Multinuclear inverse Z-grad	[11
5 mm PABB0 BB-1H/D Z-GRD Z104275/0007	[36

- 2. Select current probehead from the list by clicking on it
- 3. Click on **Define as current probe**

Figure 10.29				
	i.	No Parameterset for the current '5 mm Dual 19F/1H [04]' Do you want to create the probe	probe 's parameterset ? No	
4. Click on [	Yes			
	Edit Parameters for Prob	e: 5 mm Dual 19F/1H [04]	duct Details	
	Sample Parameters Temperature Parameters Coils Parameters Peak Power Parameters History	Probe Name: Gradient System: Part Number: Serial Number:	DUL none 2 8881 88	2 2
		ECL : nominal 1H frequency in MHz: Diameter of Probe: ATMA probe:	1 300 SB	•
		Pr	roduct Info	
		Date of Production (yyyymmdd): Production Site	2 005 05 04	
	ОК	Apply	Reset	Cancel

NOTE: On all new probeheads, most parameters are stored in a chip and are downloaded through the Pics connection. For older probeheads, fill in all the information.

5. Click on 6. Click on	OK Exit	
Figure 10.31	👹 Edit Spectrometer Parameter	R
	Preamplifier	Probe
	1H/2H 1H 1H/2H 2H XBB19F 2HS	19F 1 Inner Coll 1H 1 Outer Coll 2H 2 Outer Coll
		Save Clear cable connections Info Param Close

NOTE: If desired, the connections of the preamplifiers to the probe can be changed

7. Click on	Save
-------------	------

## 10.5 Lock File setup

#### 10.5.1 Setting the BSMS field

Sample: 0.1% Ethyl benzene in CDCl3 or any sample in CDCl3

NOTE: Do to the magnet drifting, the following procedure should be performed on a regular basis. (e.g. once a month).

1. Insert sample into the magnet

2. Type lock and select 'CDCI3'

NOTE: The system will enter the lock shift value of CDCI3 and automatically lock and adjust the lock gain.

3. Type **bsmsdisp** 

B BSMS Control	Suite			
Main Lock/Lev	/el Shim Auto	shim Service	Log Help	
AUTO		c		
Phase	Power	Gain	Lock	Shim
LOCK				
On-Off	Field	Drift	DC	
Phase	Power	Gain	Shift	
LOOP				
Gain	Time	Filter	]	
SWEEP				
On-Off	Ampl	Rate	]	
HELIUM LEVE				
Last read	Read	Measure	ן	
Absolut Differer	Prev e	ious Ac	tual Step	Reset
Sample:	down i	missing	up	
( )		10		>

- 4. Select the 'Lock/Level' tab in the BSMS Control Suite window
- 5. Switch off the lock by clicking on the LOCK 'ON/OFF' button
- 6. Click on the Lock 'Field' button
- 7. Center the lock trace within the lock window by changing the field value.



#### Figure 10.33



- 8. Press the 'Lock ON/OFF' key to lock
- 9. Shim for best resolution
- 10. Press 'Phase' button and adjust the phase for symmetry of the two lock traces
- 11. Write down the value
- 12. Type edlock at the TopSpin command line

2 Edlock										
🔳 🗋 💥 🚧	🖻 ?									
Edit 2H Lock Fi	ile				~	[Curhead 36	i: 5 mm PAE	BO BB-1H/	D Z-GRD Z1	04275/0007]
BasisFreq: 300.1	30000 [MHz]			BsmsField:	CDCI3 765	4		Nucle	us 2H	~
Solvent	Field-Cor	LockPower	LoopGain	LoopTime	LoopFilt	LockPhase	Distance	Ref	Width	Ref-Shift
Acetic	0.0	-40.0	-15.0	0.136	200	50.0	2.030	0.000	0.500	0.0000
Acetone	0.0	-40.0	-15.0	0.136	200	50.0	2.040	0.000	0.500	0.0000
CDCI3	0.0	-25.0	-15.0	0.136	200	50.0	7.240	0.000	0.500	0.0000
CD2CI2	0.0	-40.0	-15.0	0.136	200	50.0	5.320	0.000	0.500	0.0000
CD3CN	0.0	-40.0	-15.0	0.136	200	50.0	1.930	0.000	0.500	0.0000
C6D6	0.0	-26.0	-15.0	0.136	200	50.0	7.280	0.000	0.500	0.0000
D2O	0.0	-20.0	-15.0	0.136	200	50.0	4.700	0.000	0.500	0.0000
H2O+D2O	0.0	-23.0	-15.0	0.136	200	50.0	4.700	0.000	0.500	0.0000
DEE	0.0	-30.0	-15.0	0.136	200	50.0	1.070	0.000	0.500	0.0000
DME	0.0	-35.0	-15.0	0.136	200	50.0	3.300	0.000	0.500	0.0000
DMF	0.0	-25.0	-15.0	0.136	200	50.0	2.910	0.000	0.500	0.0000
DMSO	0.0	-25.0	-15.0	0.136	200	50.0	2.490	0.000	0.500	0.0000
Dioxane	0.0	-30.0	-15.0	0.136	200	50.0	3.530	0.000	0.500	0.0000
EtOD	0.0	-30.0	-15.0	0.136	200	50.0	1.110	0.000	0.500	0.0000
MeOD	40.0	-35.0	-15.0	0.136	200	50.0	3.300	0.000	0.500	0.0000
THF	0.0	-25.0	-15.0	0.136	200	50.0	1.730	0.000	0.500	0.0000
Tol	0.0	-40.0	-15.0	0.136	200	50.0	2.090	0.000	0.500	0.0000
Pyr	0.0	-25.0	-15.0	0.136	200	50.0	8.710	0.000	0.500	0.0000
CH3CN+D2O	0.0	-30.0	-15.0	0.136	200	50.0	4.700	4.700	0.500	0.0000
MeOH+D2O	0.0	-30.0	-15.0	0.136	200	50.0	4.700	4.700	0.500	0.0000
TFA	0.0	-20.0	-15.0	0.136	200	50.0	11.500	0.000	0.500	0.0000

- 10. Click on We to store the new BSMS field value
- 11. Select the first solvent in the list by clicking on it
- 12. Enter the new phase value from step 8 into the Lock Phase field
- 13. Click on 🗈 to copy the value of the selected parameter to all solvents
- 14. Click on 🔳 to save the table

15. Enter the NMR Administration password

16. Click on OK

17. Click on 🔀 to close the edlock table

#### 10.5.2 Setting the Field compensation

Sample: Tube filled with the solvent Methanol-d4

NOTE: This section describes the procedure to lock on a specific lock signal for solvents with multiple lock signals or deuterated solvents mixtures. The instructions below will guide you through the set up of successfully locking on the right solvent peak during Automation. Deuterated Methanol (CD3OD) is used in this example. To ensure an exact Field compensation value, paragraph 10.6.1. Setting the BSMS field must have been done.

- 1. Insert sample into the magnet
- 2. Type lopo MeOD at the Topspin command line

NOTE: The system will enter the lock shift value of MeOD in to the BSMS.

3. Type bsmsdisp

B BSMS Control	Suite			
Main Lock/Lev	el Shim Auto	shim Service	Log Help	
AUTO				
Phase	Power	Gain	Lock	Shim
LOCK				
On-Off	Field	Drift	DC	
Phase	Power	Gain	Shift	
LOOP				
Gain	Time	Filter		
SWEEP				
On-Off	Ampl	Rate		
HELIUM LEVEL	-			
Last read	Read	Measure		
Absoluti Differen	BY		Val Step	Reset
Sample:	down r	nissing	up	
	•	0	- O	
<		10		>

- 4. Select the 'Lock/Level' tab in the BSMS Control Suite window
- 5. Switch off the lock by clicking on the LOCK 'ON/OFF' button
- 6. Click on the Lock 'Field' button

7. Adjust the field to set the desired lock signal exactly on resonance.

NOTE: Methanol d-4 has two deuterium signals. Adjust the field for the more intense signal. Set the '**Step size**' to the lowest value to avoid loosing the lock signal.



- 8. Press the 'Lock ON/OFF' key to lock
- 9. Write down the value

10. Type edlock at the TopSpin command line

#### Figure 10.37

Figure 10.36

2 Edlock										
🔲 🗋 💥 🐆 🛙	à ?									
Edit 2H Lock Fil	le				~	[Curhead 36	: 5 mm PAB	BO BB-1H	/D Z-GRD Z1	04275/0007]
BasisFreq: 300.1	30000 (MHz)		E	IsmsField: C	DCI3 7664.	7		Nucl	eus 2H	~
Solvent	Field-Cor	LockPower	LoopGain	LoopTime	LoopFilt	LockPhase	Distance	Ref	Width	Ref-Shift
Acetic	0.0	-40.0	-15.0	0.136	200	46.5	2.030	0.000	0.500	0.0000
Acetone	0.0	-40.0	-15.0	0.136	200	46.5	2.040	0.000	0.500	0.0000
CDCI3	0.0	-25.0	-15.0	0.136	200	46.5	7.240	0.000	0.500	0.0000
CD2Cl2	0.0	-40.0	-15.0	0.136	200	46.5	5.320	0.000	0.500	0.0000
CD3CN	0.0	-40.0	-15.0	0.136	200	46.5	1.930	0.000	0.500	0.0000
C6D6	0.0	-26.0	-15.0	0.136	200	46.5	7.280	0.000	0.500	0.0000
D2O	0.0	-20.0	-15.0	0.136	200	46.5	4.700	0.000	0.500	0.0000
H2O+D2O	0.0	-23.0	-15.0	0.136	200	46.5	4.700	0.000	0.500	0.0000
DEE	0.0	-30.0	-15.0	0.136	200	46.5	1.070	0.000	0.500	0.0000
DME	0.0	-35.0	-15.0	0.136	200	46.5	3.300	0.000	0.500	0.0000
DMF	0.0	-25.0	-15.0	0.136	200	46.5	2.910	0.000	0.500	0.0000
DMSO	0.0	-25.0	-15.0	0.136	200	46.5	2.490	0.000	0.500	0.0000
Dioxane	0.0	-30.0	-15.0	0.136	200	46.5	3.530	0.000	0.500	0.0000
EtOD	0.0	-30.0	-15.0	0.136	200	46.5	1.110	0.000	0.500	0.0000
MeOD	40.0	-35.0	-15.0	0.136	200	46.5	3.300	0.000	0.500	0.0000
THF <sup>VS</sup>	0.0	-25.0	-15.0	0.136	200	46.5	1.730	0.000	0.500	0.0000
Tol	0.0	-40.0	-15.0	0.136	200	46.5	2.090	0.000	0.500	0.0000
Pyr	0.0	-25.0	-15.0	0.136	200	46.5	8.710	0.000	0.500	0.0000
CH3CN+D2O	0.0	-30.0	-15.0	0.136	200	46.5	4.700	4.700	0.500	0.0000
MeOH+D2O	0.0	-30.0	-15.0	0.136	200	46.5	4.700	4.700	0.500	0.0000
TFA	0.0	-20.0	-15.0	0.136	200	46.5	11.500	0.000	0.500	0.0000

11. Subtract the BSMS field value for CDCl3 from the field value for MeOD in step 9  $\,$ 

12. Enter the difference field value in to the 'Field compensation' for MeOD

- 13. Click on 🔳 to save the table
- 14. Enter the NMR Administration password
- 15. Click on OK



# 10.6 Observations

# 11 Hardware

### 11.1 Power up procedure for an AV-III console

The console and computer are both off.

First power the console up and just turn the IPSO unit on.

Then boot the computer. This is necessary for Windows computers so the DHCP service is started correctly. If there is no ethernet device on the router when the computer is booted, the Bruker DHCP service will not start correctly.

Once the computer is booted, and you have logged on, reset the IPSO unit so that it boots.

When the POST code gets past the stop at 'C0' and starts to load the IPSO operating system, turn the AQS, BSMS, and amplifiers on. The parts of the console that do not have ethernet connections like VT units, MAS controllers, etc, can be turned on anytime.

If you have the smaller AQS IPSO, then have to turn the AQS on to turn the IPSO on. This seems to work fine too.

When you are finished, the sync lights on all SGU/2's should be green. If not, then go into the DRU with the 'ha' screen, and reset the DRU. This will take about a minute.

Start TopSpin and do an 'ii'. If the sync led's are on for all of the SGU/2's, then you don't need to initialize the DRU again.

## 11.2 Resetting the ELCB board in the BSMS on a AV-II console

NOTE: Follow the instructions below, in case of a communication problem with the BSMS on a AV-III spectrometer do to a power glitch or during a console boot up. It is always essential in case of a BSMS problem to have stored a good shim file on a regular base.

1. Type ha on the Topspin command line

Figure 11.1

	VEB-Browser".	
Press the "Open" button	for a browser with a	
connection to this device	Э.	
Press the "Refetch addr	esses" button to relo	ad
addresses from DHCP s	erver.	
Main Operaturallan		
Main Controller		
IPSO	149.236.99.254	Open
IPSO Digital Receiver Unit-	149.236.99.254	Open
IPSO Digital Receiver Unit	149.236.99.254	Open
IPSO Digital Receiver Unit DRU1	149.236.99.254 149.236.99.89	Open
IPSO Digital Receiver Unit DRU1 Lock/Shim	149.236.99.254 149.236.99.89	Open

Figure 11.2

Lock/Shim BSMS Z100818/0936 149.236.99.20 Open

2. Click on Open

in the BSMS 'Lock/Shim' option





#### 3. Select 'Service' by clicking on it

Figure 11.4

BSMS Service Web - Windows Inter	net Explorer		
Http://149.236.99.20/serv	ice.html	Google	2
File Edit View Favorites Tools	Help		
Coople C-	T Go u 🥌 🎼 🖌 📩 Rook	marks - > O Settings - G	Spanit Lat
Coogle BSMS Service Web		l l l l l l l l l l l l l l l l l l l	( Tools +
EI	CP Comuia		1
Cher Ch	CD Servio	ce web	SER
BRUN BRUN	5 BRUN	BRUT BR	27.0
-		- <u>_</u>	
Sauker Saur	BSMS Control	Sales Sales	RES
No N	System		
Display logged messages	Log Configuration	Reset ELCB Reset KBC	~
	ER SUKE	Soft Shutdown Shims	KER
Mar M	User Level	n n	
active: user	User:	Password: Login	
Active Us	ser Configuration /	Parameters	REP
Save To Disk		1 PL PL	
	Installation Defau	lt	
CTARER CTA	Load From NVM	& CTARER C	RER
Phy Phy	BIS	BE BE	$\sim$
ELCB BIS read			-
and and	Ethomat statistic	a cha c	Ca
	Ethernet statistic	S TOWN OF U. U.	15
IP Statistics RX	IP Statistics IX	ICMP Statistics	
<u>IPIF Statistics</u>	ICP Statistics	UDP Statistics	~
CTIRER CTIV	Memory	F CTREF CT	REP
Memory Statistics	BRE	Refresh Page	$\sim$
			-
		Internet   Protected Mode: On	• 100% •

4. Click on	Reset ELCB
Figure 11.5	
	Reset ELCB Reset KBC
	Ramp Shims down, Resetting ELCB
	2017 ) 2017
Figure 11.6	Hardware ethernet addresses
	The bestware devices listed holeware he accessed
	and configured with a "WEB-Browser".
	Brace the "Open" butten for a browcer with a

to this device			
Refetch addre	esses" butto	on to reload	d
rom DHCP se	erver.		
oller			
	149.236.9	99.254	Oper
eiver Unit			
	149.236.9	99.89	Oper
	and the second of the second o		-
	rom DHCP se oller eiver Unit	rom DHCP server. oller 149.236.9 eiver Unit 149.236.9	rom DHCP server. oller 149.236.99.254 eiver Unit 149.236.99.89

5. Click on Close

6. Type rsh <Name of the last good shim file>

## 11.3 Downloading a new DRU Firmware

NOTE: The instructions below are intended for installing a new DRU Firmware in case from a request by Bruker Center or the Application Hotline to fix an problem with the Digital Receiver unit. You would be instructed to down load the Firmware from a ftp site. If a a message to install a new DRU firmware pops up during a TopSpin software upgrade, follow the instructions showing in the message box.

1. Type ha on the Topspin command line

Figure 11.7

Hardware ethernet addres	sses	×
The hardware devices lis and configured with a "W	sted below can be a VEB-Browser".	ccessed
Press the "Open" button connection to this device	i for a browser with a e.	ļ,
Press the "Refetch addr addresses from DHCP si	esses" button to relo erver.	ad
Main Controller		
IPSO	149.236.99.254	Open
∣ Digital Receiver Unit —		
DRU1	149.236.99.89	Open
Lock/Shim		
BSMS Z100818/0936	149.236.99.20	Open
Refetch ac	ddresses Print	Close

Figure 11.8

Digital Receiver Unit		
DRU1	149.236.99.89	Open

2. Click on Open in the DRU1 'Digital Receiver Unit' option





3. Select 'AQS Firmware Setup' by clicking on it



4. Select 'Load new DRU Firmware' by clicking on it

#### Figure 11.11



NOTE: The current DRU Firmware version is displayed in this window. Check if the new Firmware has a newer date, then proceed with the steps below. If the new Firmware has the same or older date, no further action is necessary and the Web window can be closed.



6. Select the Firmware file

Click on install firm	ware	
re 11.13		
Firmware Download Status	- Windows Internet Explorer	
😋 💽 👻 🙋 http://149.236	393.89/DownldFirmStat.html	2
File Edit View Favorites	Tools Help	
Links 🖉 Microsoft Windows Up	date 🖕 Bruker BioSpin 🧑 Contact HP Worldwide 🧏 Adobe - Adobe Reader downloa	ad I 🗲 Smaall 🐖
		Snagit E
🎽 🍄 🔡 🔻 🍎 AQS Ma	in 🔰 🚺 Firmware Download S 🗙 🔤 🕤 🐨 🗟 👻 🖶 👻 Pa	age 👻 🌍 Tools 🤊
BRUKER	DEB Service Web Firmware Download Status	
BRUKER	<b>DEB Service Web</b> Firmware Download Status	A DIRER
Action	DEB Service Web Firmware Download Status Details	Status
Action File Download	DEB Service Web Firmware Download Status Details Filename: dru_firmware_080509.gz	Status OK
Action File Download File Verification	DEB Service Web Firmware Download Status	Status OK OK
Action File Download File Verification Firmware Storing	DEB Service Web Firmware Download Status Details Filename: dru_firmware_080509.gz Checking Checksum Erasing the FLASH and reprogramming the firmware	Status OK OK WAIT
Action File Download File Verification Firmware Storing Firmware Checking	DEB Service Web Firmware Download Status Details Filename: dru_firmware_080509.gz Checking Checksum Erasing the FLASH and reprogramming the firmware Verifying the programmed data	Status OK OK WAIT
Action File Download File Verification Firmware Storing Firmware Checking End	DEB Service Web Firmware Download Status Details Filename: dru_firmware_080509.gz Checking Checksum Erasing the FLASH and reprogramming the firmware Verifying the programmed data The program is loaded. Resetting the Board!	Status OK OK WAIT
Action File Download File Verification Firmware Storing Firmware Checking End	DEB Service Web Firmware Download Status Details Filename: dru_firmware_080509.gz Checking Checksum Erasing the FLASH and reprogramming the firmware Verifying the programmed data The program is loaded. Resetting the Board!	Status OK OK WAIT

NOTE: After the program is loaded and the board is reset the current Web page in Figure 11.13 will close automatically and the Web page in Figure 11.14 is displayed.

#### Figure 11.14



- 9. Click on 🔀 to close the Web page
- Figure 11.15

and configured with a "	listed below can be acc WEB_Browser"	cessed
and configured with a	WEB-Browser .	
Press the "Open" butto	on for a browser with a	
Press the "Refetch add	ue. dresses" button to reloa	nd
addresses from DHCP	server.	
Main Controller		
Main Controller	149.236.99.254	Open
Main Controller IPSO Digital Receiver Unit—	149.236.99.254	Open
Main Controller IPSO Digital Receiver Unit DRU1	149.236.99.254 149.236.99.89	Open
Main Controller IPSO Digital Receiver Unit DRU1 Lock/Shim	149.236.99.254 149.236.99.89	Open
Main Controller IPSO Digital Receiver Unit DRU1 Lock/Shim BSMS Z100818/0936	149.236.99.254 149.236.99.89	Open

10. Click on Close

# 11.4 Observations

# Appendix

Α

### A.1 Standard Parameter set list

N AL27ND - 27Al exp. no decoupling N B11ZG - 11B exp. no decoupling N C13APT - Attached Proton Test using jmod pulse program N C13CPD - C13 exp. comp. pulse dec. 1024 scans N C13CPD32 - C13 exp. comp. pulse dec. 32 scans N C13CPDSN - C13 exp. comp. pulse dec. with signal-to-noise calc. N C13DE45SN - C13 dept all positive with signal-to-noise calc. N C13DEPT45 - C13 dept all positive N C13DEPT90 - C13 dept CH-only N C13DEPT135 - C13 dept CH, CH3 pos. CH2 neg. N C13DEPT135p - dept135 with phase of previous C13 N C13GD - C13 exp. gated decoupling N C13IG - C13 exp. inverse gated decoupling N C13MULT - 13C automatic multiplicity determination N C130FF - C13 exp. off resonance N C13PPTI - C13 exp. with peak picking in title N C13HUMP - 13C hump (lineshape) test N C13RESOL - 13C resolution (half width) test N C13SENS - 13C sensitivity (SINO) test N CD111ZG - 111Cd exp. no decoupling N CD113ZG - 113Cd exp. no decoupling N CL35ZG - 35Cl exp. no decoupling N CL37ZG - 37Cl exp. no decoupling N F19 - 19F exp. no decoupling N F19CPD - 19F exp. comp. pulse decoupling N GA71ZG - 71Ga exp. no decoupling N HG199CPD - 199Hg exp. comp. pulse decoupling N HMQC1D - 1D version of the HMQC N LC1D12 - 1H, double presaturation N LC1DCWPS - 1H, multiple presaturation N LC1DWTDC - 1H, mult. WET suppr., 13C decoupling N LCMLCWPS - TOCSY TPPI, mult. presat., 13C decoupling N N15 - 15N exp. no decoupling N N15IG - 15N exp. inverse gated N N15INEPT - 15N exp. inept N NA23ZG - 23Na exp. no decoupling N NOEDIFF - 1H noe difference N 017ZG - 170 exp. no decoupling N P31 - 31P exp. no decoupling N P31CPD - 31P exp. comp. pulse decoupling N PROB11DEC - 1H with B11 decoupling

```
N PROF19DEC - 1H with F19 decoupling
N PROP31DEC - 1H with P31 decoupling
N PROTON - 1H experiment 16 scans
N PROTON128 - 1H experiment 128 scans
N PROTONinfo - 1H experiment with info table
N PROTONCONLF - 1H exp. with conditional low field plot
N PROTONEXP - 1H experiment + expansions
N PROTONLF - 1H experiment + low field plot
N PROTONLFEXP - 1H experiment + low field plot + expansions
N PROTONNR - 1H exp. non spinning
N PROTONNREXP - 1H exp. non spinning + expansions
N PROTONNRLF - 1H exp. non spinning + low field plot
N PRONRLFEXP - 1H exp. non spinning + low field plot + expansions
N PROHOMODEC - 1H homo decoupling experiment
N PROTONT1 - 1H T1 Relaxation measurement
N PROHUMP - 1H hump (lineshape) test
N PRORESOL - 1H resolution (half width) test
N PROSENS - 1H sensitivity (SINO) test
N PT195ZG - 195Pt exp. no decoupling
N RH103ZG - 103Rh exp. no decoupling
N SE77ZG - 77Se exp. no decoupling
N SELCO1H - 1D COSY using sel. excitation w/a shaped pulse
N SELMLZF1H - 1D homo. Hartman-Hahn transfer using MLEV17 and
sel. exc. w/a shaped pulse
N SELNO1H - 1D NOESY using sel. exc. w/a shaped pulse
N SELRO1H - 1D ROESY using sel. exc. w/a shaped pulse
N SELZG1H - 1D sequence using sel. exc. w/a shaped pulse
N SI29IG - 29Si exp. inverse gated decoupling
N SN119IG - 119Sn exp. inverse gated decoupling
N WATERSUP - 1H water supression test
N WATER - water supression
C COSY45SW - sw opt. COSY45 (magn. mode)
C COSY90SW - sw opt. COSY90 (magn. mode)
C COSYDQFPHSW - sw opt. COSY with dq filter (States-TPPI)
C COSYGPDFPHSW - sw opt. COSY with gradients and dq filter
(States-TPPI)
C COSYGPMFSW - sw opt. COSY with gradients and mq filter (magn.
mode)
C COSYGPSW - sw opt. COSY with gradients (magn. mode)
C HCCOSW - sw opt. CH-correlation
C HCCOLOCSW - sw opt. COLOC
C INV4SW - sw opt. HMQC (magn. mode)
C INV4PHSW - sw opt. HMQC (States-TPPI)
C INV4GPSW - sw opt. HMQC with gradients (magn. mode)
C INV4GPMLSW - sw opt. HMQC-TOCSY with gradients (magn. mode)
C INVBSW - sw opt. HMQC using BIRD pulse (magn. mode)
C INVBPHSW - sw opt. HMQC using BIRD pulse (States-TPPI)
C INV4GPLPLRNDSW- - HMBC with gradients and low pass J-filter
```

- C INV4GPLRNDSW sw opt. HMBC with gradients
- C INV4LPLRNDSW sw opt. HMBC with low pass J-filter (magn. mode)
- C INVIGPMLPHSW sw opt. HSQC-TOCSY with gradients (States-TPPI)
- C INVIGPPHSW sw opt. HSQC with gradients (States-TPPI)
- C MLEVPHSW sw opt. TOCSY (States-TPPI)
- C NOESYPHSW sw opt. NOESY (States-TPPI)
- C ROESYPHSW sw opt. ROESY (States-TPPI)
- C INVIETGPSW sw opt. HSQC with gradients (e/a TPPI)

C INVIETGPSISW - sw opt. HSQC sens. improved with gradients (e/a TPPI)

- C INVIETGPMLSW sw opt. HSQC-TOCSY with gradients (e/a TPPI)
- C INVIEDETGPSW sw opt. edited HSQC with gradients (e/a TPPI)
- C INVIEDGPPHSW sw opt. edited HSQC with gradients (States-TPPI)

## A.2 Standard Test Samples

#### 1H Lineshape

0.3% Chloroform in Acetone-d6 (CRYO-probes)1% Chloroform in Acetone-d6 (500MHz and up)3% Chloroform in Acetone-d6 (up to 500MHz)

#### **1H Sensitivity**

0.1% Ethyl benzene in Chloroform-d1H Solvent Suppression2mM Sucrose in 90% H2O, 10% D2O2mM Lisozyme in 90% H2O, 10% D2O

#### **13C Sensitivity**

10% Ethyl benzene in Chloroform-d 40% p-Dioxane in 60% Benzene-d6

#### 31P Sensitivity

Triphenylphosphate in Chloroform-d

#### 15N Sensitivity

90% Formamide in Dimethyl Sulfoxide-d6

#### Calibration of the 13C and 15N 90 degree pulses

0.1M 15N-Urea, 0.1M 13C-Methanol in Dimethyl Sulfoxide-d6

19F Sensitivity Trifluorotoluene in Chloroform-d

#### **Temperature Calibration**

80% Ethylene Glycol in Dimethyl Sulfoxide-d6 (High Temperature)4% Methanol in 96% Methanol-d (Low Temperature)

#### 1D and 2D Experiments

100mg/mL Cholesteryl Acetate in Chloroform-d10mg Strychnine in Chloroform-d50mM Gramicidine in Dimethyl Sulfoxide-d6
## **B** Contact

For further technical assistance, please do not hesitate to contact your nearest BRUKER dealer or contact us directly at:

BRUKER BioSpin Corporation 15 Fortune Drive, Manning Park Billerica, MA 01821 USA

Phone: 978) 667-9580 Ext. 5444 FAX: 978) 667-2955 Email: applab@bruker-biospin.com Internet: www.bruker-biospin.com

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info@bruker-biospin.com www.bruker-biospin.com