Console Acceptance Tests

MERCURYplus NMR Spectrometer Systems with VnmrJ

Pub. No. 01-999255-00, Rev. B0904

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Applicability of manual:
MERCURYplus NMR spectrometer systems running VnmrJ

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A0604 – Initial release with VnmrJ 1.1D
A0804 – Corrections and updates
B0904 – Shipping kit correction ECO

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SAFETY PRECAUTIONS

The following warning and caution notices illustrate the style used in Varian manuals for safety precaution notices and explain when each type is used:

This symbol might be used on warning labels attached to the equipment. When you see this symbol, refer to the relevant manual for the information referred to by the warning label.

WARNING: ***Warnings* are used when failure to observe instructions or precautions could result in injury or death to humans or animals, or significant property damage.**

CAUTION: ***Cautions* are used when failure to observe instructions could result in serious damage to equipment or loss of data.**

Warning Notices

Observe the following precautions during installation, operation, maintenance, and repair of the instrument. Failure to comply with these warnings, or with specific warnings elsewhere in Varian manuals, violates safety standards of design, manufacture, and intended use of the instrument. Varian assumes no liability for customer failure to comply with these precautions.

WARNING: **Persons with implanted or attached medical devices such as pacemakers and prosthetic parts must remain outside the 5-gauss perimeter from the centerline of the magnet.**

The superconducting magnet system generates strong magnetic fields that can affect operation of some cardiac pacemakers or harm implanted or attached devices such as prosthetic parts and metal blood vessel clips and clamps.

Pacemaker wearers should consult the user manual provided by the pacemaker manufacturer or contact the pacemaker manufacturer to determine the effect on a specific pacemaker. Pacemaker wearers should also always notify their physician and discuss the health risks of being in proximity to magnetic fields. Wearers of metal prosthetics and implants should contact their physician to determine if a danger exists.

Refer to the manuals supplied with the magnet for the size of a typical 5-gauss stray field. This gauss level should be checked after the magnet is installed.

WARNING: **Keep metal objects outside the 10-gauss perimeter from the centerline of the magnet.**

The strong magnetic field surrounding the magnet attracts objects containing steel, iron, or other ferromagnetic materials, which includes most ordinary tools, electronic equipment, compressed gas cylinders, steel chairs, and steel carts. Unless restrained, such objects can suddenly fly towards the magnet, causing possible personal injury and extensive damage to the probe, dewar, and superconducting solenoid. The greater the mass of the object, the more the magnet attracts the object.

Only non ferromagnetic materials—plastics, aluminum, wood, nonmagnetic stainless steel, etc.—should be used in the area around the magnet. If an object

is stuck to the magnet surface and cannot easily be removed by hand, contact Varian service for assistance.

Refer to the manuals supplied with the magnet for the size of a typical 10-gauss stray field. This gauss level should be checked after the magnet is installed.

WARNING: **Only qualified maintenance personnel shall remove equipment covers or make internal adjustments.**

Dangerous high voltages that can kill or injure exist inside the instrument. Before working inside a cabinet, turn off the main system power switch located on the back of the console.

WARNING: **Do not substitute parts or modify the instrument.**

Any unauthorized modification could injure personnel or damage equipment and potentially terminate the warranty agreements and/or service contract. Written authorization approved by a Varian, Inc. product manager is required to implement any changes to the hardware of a Varian NMR spectrometer. Maintain safety features by referring system service to a Varian service office.

WARNING: **Do not operate in the presence of flammable gases or fumes.**

Operation with flammable gases or fumes present creates the risk of injury or death from toxic fumes, explosion, or fire.

WARNING: **Leave area immediately in the event of a magnet quench.**

If the magnet should quench (sudden appearance of gasses from the top of the dewar), leave the area immediately. Sudden release of helium or nitrogen gases can rapidly displace oxygen in an enclosed space creating a possibility of asphyxiation. Helium will displace air from the top of a room and cold nitrogen can displace air from the lower levels of a room. Do not return until the oxygen level returns to normal.

WARNING: **Avoid helium or nitrogen contact with any part of the body.**

Cold gasses or liquids (helium and nitrogen) contacting the body can cause an injury similar to a burn. Never place your head over the helium and nitrogen exit tubes on top of the magnet. If cold gasses or liquids contact the body, seek immediate medical attention, especially if the skin is blistered or the eyes are affected.

WARNING: **Do not look down the upper barrel.**

Unless the probe is removed from the magnet, never look down the upper barrel. You could be injured by the sample tube as it ejects pneumatically from the probe.

WARNING: **Do not exceed the boiling or freezing point of a sample during variable temperature experiments.**

A sample tube subjected to a change in temperature can build up excessive pressure, which can break the sample tube glass and cause injury by flying glass and toxic materials. To avoid this hazard, establish the freezing and boiling point of a sample before doing a variable temperature experiment.

WARNING: **Support the magnet and prevent it from tipping over.**

The magnet dewar has a high center of gravity and could tip over in an earthquake or after being struck by a large object, injuring personnel and causing sudden, dangerous release of nitrogen and helium gasses from the dewar. Therefore, the magnet must be supported by at least one of two methods: with ropes suspended from the ceiling or with the antivibration legs bolted to the floor. Refer to the *Installation Planning Manual* for details.

WARNING: **Do not remove the relief valves on the vent tubes.**

The relief valves prevent air from entering the nitrogen and helium vent tubes. Air that enters the magnet contains moisture that can freeze, causing blockage of the vent tubes and possibly extensive damage to the magnet. It could also cause a sudden dangerous release of nitrogen and helium gases from the dewar. Except when transferring nitrogen or helium, be certain that the relief valves are secured on the vent tubes.

WARNING: **On magnets with removable quench tubes, keep the tubes in place except during helium servicing.**

On Varian 200- and 300-MHz 54-mm magnets only, the dewar includes removable helium vent tubes. If the magnet dewar should quench (sudden appearance of gases from the top of the dewar) and the vent tubes are not in place, the helium gas would be partially vented sideways, possibly injuring the skin and eyes of personnel beside the magnet. During helium servicing, when the tubes must be removed, follow carefully the instructions and safety precautions given in the manual supplied with the magnet.

Caution Notices

Observe the following precautions during installation, operation, maintenance, and repair of the instrument. Failure to comply with these cautions, or with specific cautions elsewhere in Varian manuals, violates safety standards of design, manufacture, and intended use of the instrument. Varian assumes no liability for customer failure to comply with these precautions.

CAUTION: **Keep magnetic media, ATM and credit cards, and watches outside the 5-gauss perimeter from the centerline of the magnet.**

The strong magnetic field surrounding a superconducting magnet can erase magnetic media such as floppy disks and tapes. The field can also damage the strip of magnetic media found on credit cards, automatic teller machine (ATM) cards, and similar plastic cards. Many wrist and pocket watches are also susceptible to damage from intense magnetism.

Refer to the manuals supplied with the magnet for the size of a typical 5-gauss stray field. This gauss level should be checked after the magnet is installed.

CAUTION: **Keep the PCs, (including the LC STAR workstation) beyond the 5-gauss perimeter of the magnet.**

Avoid equipment damage or data loss by keeping PCs (including the LC workstation PC) well away from the magnet. Generally, keep the PC beyond

the 5-gauss perimeter of the magnet. Refer to the *Installation Planning Guide* for magnet field plots.

CAUTION: **Check helium and nitrogen gas flow meters daily.**

Record the readings to establish the operating level. The readings will vary somewhat because of changes in barometric pressure from weather fronts. If the readings for either gas should change abruptly, contact qualified maintenance personnel. Failure to correct the cause of abnormal readings could result in extensive equipment damage.

CAUTION: **Never operate solids high-power amplifiers with liquids probes.**

On systems with solids high-power amplifiers, never operate the amplifiers with a liquids probe. The high power available from these amplifiers will destroy liquids probes. Use the appropriate high-power probe with the high-power amplifier.

CAUTION: **Take electrostatic discharge (ESD) precautions to avoid damage to sensitive electronic components.**

Wear grounded antistatic wristband or equivalent before touching any parts inside the doors and covers of the spectrometer system. Also, take ESD precautions when working near the exposed cable connectors on the back of the console.

Radio-Frequency Emission Regulations

The covers on the instrument form a barrier to radio-frequency (rf) energy. Removing any of the covers or modifying the instrument may lead to increased susceptibility to rf interference within the instrument and may increase the rf energy transmitted by the instrument in violation of regulations covering rf emissions. It is the operator's responsibility to maintain the instrument in a condition that does not violate rf emission requirements.

Chapter 1. Introduction

Sections in this chapter:

Following each installation of a Varian, Inc. *MERCURYplus* NMR spectrometer system, an installation engineer tests and demonstrates the instrument’s operation using the procedures in this manual.

The following is an overview of the chapters in this manual:

- Chapter 2, “Console and Magnet Test Procedures,” provides the acceptance test procedures.
- Chapter 3, “Consoles and Magnets Specifications,” provides the acceptance test specifications.
- Chapter 4, “Customer Training,” provides basic spectrometer maintenance and operation training.
- Chapter 5, “Acceptance Test Results,” contains forms for recording test results.

The acceptance test procedures and specifications for each probe are contained in a separate probe family manual, for example procedures and specifications for AutoSwitchable probes are contained in the *AutoSwitchable NMR Probes* manual.

Only the lineshape and signal-to-noise tests are preformed manually. All other probe calibrations are performed by the instrument during the AutoCalibration procedures.

1.1 Overview of the Acceptance Testing Process

The objectives of the acceptance tests procedures are threefold:

- To identify the tests to be performed during system installation.
- To identify the precise methods by which these tests are performed.
- To leave the instrument in a calibrated, ready to use, state.

Acceptance Tests

Acceptance test procedures are arranged by the type of specification. These procedures cover the basic specifications of the instrument—signal-to-noise, resolution, and lineshape—and are not intended to reflect the full range of operating capabilities or features of a *MERCURYplus* NMR spectrometer. The order in which the tests are performed is determined by the installation engineer. Some procedures use results from other procedures

Performance of additional tests beyond those described in this manual must be agreed upon in writing as part of the customer contract.

Acceptance Specifications

All specifications are subject to change without notice. The specifications published in this manual shall prevail unless negotiation or customer contract determines otherwise. Refer to the text in each chapter for other conditions.

Request for additional specifications beyond those listed in this manual must be agreed upon in writing as part of the customer contract. The following policies are in effect at installation:

- **Specifications Policy for Probes Used in Systems other than *MERCURYplus*** – No guarantee is given that probes purchased for use in systems other than *MERCURYplus* will meet current specifications.
- **Testing Policy for Indirect Detection Probes used for Direct Observe Broadband Performance** – Probes designed for indirect detection applications are tested for indirect detection performance only. Indirect detection acceptance tests are performed only if an indirect detection probe was purchased for use with the *MERCURYplus*.
- **Sample Tubes Policy** – Tests are performed in 5-mm sample tubes with 0.38 mm wall thickness (Wilmad 528-PP, or equivalent) and 10-mm sample tubes with 0.46 mm wall thickness (Wilmad 513-7PP, or equivalent). Using sample tubes with thinner walls (Wilmad 5-mm 545-PPT, or equivalent; Wilmad 10-mm 513-7PPT, or equivalent) increases signal-to-noise.

Computer Audit

A computer audit form is included in “Computer Audit,” page 61. The information from this form will help Varian, Inc. assist you better in distributing future software upgrades and avoiding hardware compatibility problems. You are asked for information about all computers directly connected to the spectrometer or else used to process NMR data.

Installation Checklist

An installation checklist form is included in “System Installation Checklist,” page 65.

System Documentation Review

Following the completion of the acceptance tests and computer audit, the installation engineer will review the following system documentation with the customer:

- Software Object Code License Agreement.
- Varian, Inc. and OEM manuals.
- Warranty coverage and where to telephone for information.

Basic System Demonstration

The installation engineer will also demonstrate the basic operation of the system to the laboratory staff. The objective of the demonstration is to familiarize the customer with system features and safety requirements, as well as to assure that all mechanical and electrical functions are operating properly.

The system demonstration includes the following items:

Magnet Demonstration

The following are demonstrated:

- Posting requirements for magnetic field warning signs.
- Cryogenics handling procedures and safety precautions.
- Magnet refilling.
- Flowmeters.
- Homogeneity disturbances.

Console and Probe Demonstration

The following are demonstrated:

- Loading programs (VnmrJ, Optional VnmrJ packages, operating system (OS)).
- Experiment setup, including installing the probe in the magnet.
- Basic instrument operation to obtain typical spectra, including probe tuning, magnet homogeneity shimming, and printer/plotter operation. (Note that Varian, Inc. installation engineers are not responsible for, or trained to, run any spectra not described in this manual.)
- AutoCalibration of key probe parameters such as ^{1}H pw90, ^{13}C pwx90, decoupler field, gradient strength (if gradients are present), and other probe specific parameters.
- Demonstration of automated data acquisition. Using the 2-Ethyl-1-indanone sample provided with the console the following experiments will be run:
  - 1D Experiments: ^{1}H, ^{13}C{^{1}H}, APT and DEPT.
  - 2D Non gradient experiments: NOESY and TOCSY (for non-gradient system or probe COSY is also run).
  - 2D Gradient experiments: gCOSY, gHSQC, gHMBC will be demonstrated if gradients are present.
- Walk through the demonstration spectra and the “Interpreting the Calibration and Indanone Spectra,” page 46.
- Demonstration of gradient shimming using PFG gradients, if present, or homospoil.
- Review how to use the manuals (online and hard copy) and where to find information.
- Review the post installation 30 day check list.

Detailed specifications and circuit descriptions are not covered.

Formal training in the operation and maintenance of the spectrometer is conducted by Varian, Inc. at periodically scheduled training seminars held in most Varian, Inc. NMR Application Laboratories. Installation engineers are responsible for guiding you through the acquisition of the spectra as provided in the manual. The installation engineer is not responsible for interpretation of the results beyond what is provided in this manual. On-site training is available in some geographic locations. Contact your sales representative (4.9 “Warranty and Who to Call for Assistance” page 57) for further information on availability and pricing for these courses.

To make the system demonstration most beneficial, the customer should review Varian, Inc. and OEM manuals before viewing the demonstration.

1.2 General Acceptance Testing Requirements

Each *MERCURYplus* spectrometer is designed to provide high-resolution performance when operated in an environment as specified in the *Installation Planning Guide*. Unless both the specific requirements of this manual and the general requirements specified in the *MERCURYplus Installation Planning Guide* are met, Varian, Inc. cannot warrant that the NMR spectrometer system will meet the published specifications.

1.3 Samples Required for Acceptance Tests

The *MERCURYplus* spectrometer is supplied with the samples listed in Table 1

Table 1. Samples Required for Console Acceptance Tests

Test Sample	*Sample Tube (mm)*	*Nucleus*	*Sample Part Number*
autotest sample; 0.1% ^{13}C enriched methanol in 1% H_2O/99% D_2O	5	^{1}H and ^{13}C	00-968120-68
^{13}C enriched 1% methyl iodide, 1% trimethyl phosphite, and 0.2% Cr(AcAc) in chloroform-d	5	^{1}H, ^{13}C and ^{31}P	00-968120-96
doped 4-Hz H_2O/D_2O (0.1 mg/ml $GdCl_3$ in 1% H_2O in D_2O)	5	^{1}H	01-901855-01
2% 2-ethyl-1-indanone in chloroform-d	5	^{1}H and ^{13}C	01-901855-03
0.1% ethylbenzene, 0.01% TMS, 99.89% deuterochloroform ($CDCl_3$)	5	^{1}H	00-968120-70
0.1% ethylbenzene, 0.01% TMS, 99.89% deuterochloroform ($CDCl_3$)	10	^{1}H	00-968123-70
chloroform in acetone-d_6 lineshape	5	^{1}H	00-968120-xx
100% methanol (reagent grade) –50 to +25 (Low)	5	^{1}H	00-968120-80
100% ethylene glycol (reagent grade) +25 to +100 (High)	5	^{1}H	00-968120-79

The samples required for acceptance testing of any individual probe are supplied with the probe. Typical samples required for probe acceptance tests are: chloroform in acetone-d_6, ethyl benzene in chloroform-d, and ASTM (40% p-dioxane in 60% benzene-d6). Other samples might be required depending upon the type of probe. The specific sample requirements and test procedures are specified in the manual for each probe.

1.4 General Testing and Specification Requirements

- The *MERCURYplus* performance specifications in effect at the time of your order are used to evaluate the system.
- The appropriate quarter-wavelength cable must be used for each nucleus except AutoSwitchable probes operated in 4-nucleus mode. The ^{31}P quarter-wavelength cable is used in this case.
- Homogeneity settings must be optimized for each sample (manual shimming may be required in any or all cases). The shim parameters for resolution tests on each probe should be recorded in a log book and in a separate file name (in the directory `/vnmr/shims`) for each probe. For example, for a 5-mm switchable probe, the shim parameters can be saved with the command `svs('sw5res')`. These values can then

be used as a starting point when adjusting the homogeneity on unknown samples, by the command `rts('sw5res')`.

- The probe must be tuned to the appropriate frequency.
- Spinning speed must be set to the following:

Sample (mm)	*Nuclei*	*Speed (Hz)*
5	all	20–26
10	all	15

Spinning 10-mm tubes faster than 15 Hz can cause vortexing in samples, severely degrading the resolution.

- Some test parameters are stored in the disk library `/vnmr/tests`. To see the parameter sets available for the standard tests, use the VnmrJ File Browser. Other sets come from `/vnmr/stdpar`.
- Many of the probe parameters and performance specifications will be determined automatically and the corresponding manual tests will NOT be run by the installer. Certain tests, such as signal-to-noise and lineshape will be run manually. Tests corresponding to the automatic performance tests are provided should you want to run them at a later time.
- For all sensitivity tests, the value of `pw` must be changed to the value of the 90° pulse found in the pulse width test on the same probe and nucleus.
- During calibration, VnmrJ creates an appropriate pw array to determine the 90° pulse width. For manually run observe pulse width tests, an appropriate array of `pw` values must be entered to determine the 360° pulse. The 360° pulse is the second non-zero pulse that gives minimum intensity of the spectrum. The 360° pulse is usually determined by interpolation between a value that gives a positive signal, and a value that gives a negative signal. The 90° pulse width is one quarter the 360° pulse (360/4).
- Signal-to-noise (S/N) is measured by the computer as follows:

$$\text{S/N} = \frac{\text{maximum amplitude of peak}}{2 \times \text{root mean square of noise region}}$$

- Lineshape should be measured digitally with the aid of the system software. The properly scaled spectra should also be plotted and the plot retained.
- Digital determination of lineshape:
  1. Display and expand the desired peak.
  2. Enter **`nm`**, then **`dc`** for drift correction to ensure a flat baseline. Set **`vs=10000`**. Click  icon to display the horizontal threshold cursor. Set **`th=55`** (the 0.55% level).
  3. Click the  icon, two vertical cursors are displayed. Align the cursors on the intersections of the horizontal cursor and the peak. Enter **`delta?`** to see the difference in Hz between the cursors.
  4. Set **`th=11`** (the 0.11% level) and repeat.
  5. You can also use the macro `res`. Place the cursor on the resonance of interest and type `res` on the command line.
- Determination of lineshape from a plot:

1. Use a large enough plot width to allow accurate determination of the baseline. The baseline should be drawn through the center of the noise, in a region of the spectrum with no peaks.
2. The 0.55% and 0.11% levels are then measured from the baseline and calculated from the height of the peak and the value of `vs`. For example, if a peak is 9.0 cm high with `vs=200`, then the 0.55% level on a 100-fold vertical expansion (`vs=20000`) is 9×0.55, or 4.95 cm from the baseline.

If the noise is significant at the 0.55% and 0.11% levels, the linewidth should be measured horizontally to the center of the noise.

- Use the `dsnmax` macro to determine signal-to-noise (sensitivity). You can also choose a noise region free of any anomalous features with the cursors. Neither cursor should be placed any closer to an edge of the spectrum than 10 percent of the value of `sw`. This should produce the best possible signal-to-noise that is representative of the spectrum.
- The results of all tests should be plotted to create a permanent record. Include a descriptive label and a list of parameters. These plots can then be saved as part of the acceptance tests documentation.

Chapter 2. Console and Magnet Test Procedures

Sections in this chapter:

This chapter contains the procedures for testing and demonstrate the operation of the NMR consoles and magnets. Refer to Chapter 3, “Consoles and Magnets Specifications,” and record results in Chapter 5, “Acceptance Test Results,” using the provided forms.

2.1 Calibrating a Probe

This procedure uses the AutoCalibration features of the VnmrJ Experimental interface.

Lineshape and resolution tests described in the probe manual shipped with your probe must be run before these procedures are run. The probe calibration procedures create probe calibration files that are used for some of the console procedures. The probe calibration data written to these probe files will meet or exceed the specifications for the probe. Probe performance tests and calibrations that are executed during AutoCalibration will not be repeated manually.

Probe Calibration Samples

The following samples are required for probe calibration:

Sample	*Sample Tube (mm)*	*Nuclei*	*Part No.*
^{13}C enriched 1% methyl iodide, 1% trimethyl phosphite, and 0.2% Cr(AcAc) in chloroform-d	5	^{1}H, ^{13}C and ^{31}P	00-968120-96
doped 4-Hz H_2O/D_2O (0.1 mg/ml $GdCl_3$ in 1% H_2O in D_2O)	5	^{1}H	01-901855-01

Before You Start

Before beginning the probe calibration you should already have the following:

- defined a printer and plotter (see *VnmrJ Installation and Administrator* for details)
- obtained the lineshape specifications (see the appropriate probe manual for details).

1. Log in as the VnmrJ administrator.
2. Click the **Locator Statements** menu ( ) and select **System Param Files** from under the **Sort NMR Parameter Files** category.
3. Click the title above the right most column and select **Directory** from the lists that is displayed.
4. Select **`shmd2o`** and drag it to the VnmrJ graphics canvas.
5. Insert the **doped 2-Hz H_2O/D_2O** sample (01-901855-01).
6. Go to the **Lock page** (Start tab -> Lock page). Click **Lock Scan** and set **Lock** to **Off**.
7. Adjust **Z0** to be on resonance. Adjust this as close as possible; the shimmap depends on accuracy.
8. Adjust **Power**, **Gain**, and **Phase** for a steady lock level of about **80**.
9. Click **Lock Scan** to stop the lock update. Click Standard to go to the **Study** page.

Setting Up the Probe Calibration File

Before you calibrate a probe for the first time, you must set up the probe calibration file. You must be logged in as the VnmrJ administrator for this procedure.

1. Select **Utilities** -> **Standard Calibration Experiments** -> **Calibrate Probe**, or click the **Probe** button on the hardware bar, see Figure 1.
2. Click the **Edit Probe** box — editing options appear in the Probe window.

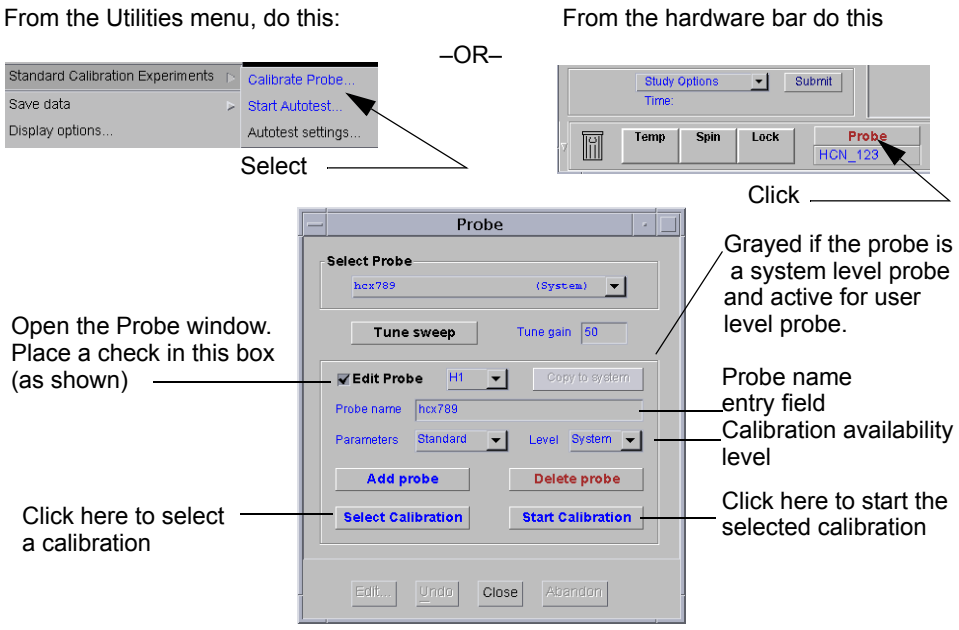
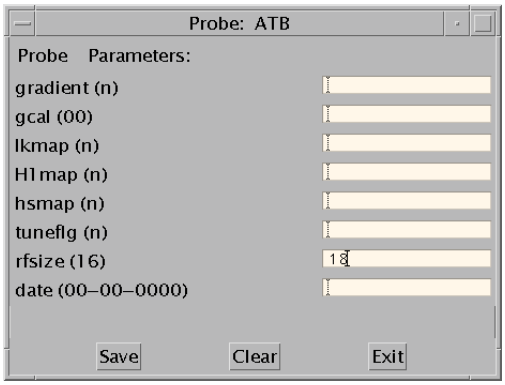


Figure 1. Calibrating a Probe

3. Enter the name of the probe in the **Probe name** field (e.g. asw_5mm).
4. Select **System** from the **Level** drop down menu.

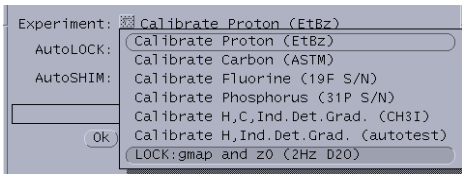
   The *System* selection writes the calibrations into: `/vnmr/probes/probe_name` and makes all calibration available to all users. The *User* selection writes the calibrations into:`~/vnmrsys/probes/probe_name` and are available only to the logged in user creating the calibration file.
5. Leave **Parameters** at **zero**.
6. Click the **Add probe**.
7. From the menu next to **Edit Probe**, select **Probe**. Enter the correct value for **rfsize**, click **Save**, **Exit**. Refer to the probe manual for the correct value.



Calibrating Z0 and Make LOCK gmap

This procedure calibrates Z0 and makes a gradient map for gradient shimming for systems with gradients and gradient probes. Gradient shimming will be done for non-gradient systems by using homospoil.

1. Click the **Select Calibration** button in the Probe window.
2. Set **AutoLOCK** and **AutoSHIM** to **NO**.
3. Right click the **Experiment** drop-down menu and select **Lock:gmap and z0 (4-Hz D_2O)** from the list of calibration options.

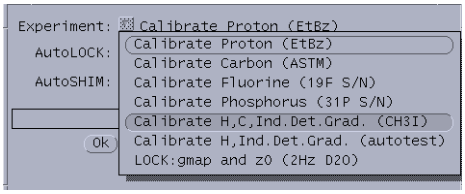


4. Click **Ok**.
5. Click **Exit**.
6. The message `Set z0 exactly on-resonance before starting acquisition` is displayed. Refer to the *VnmrJ Liquids NMR* manual for more information on setting the lock.
7. Click **Confirm** in the popup prompt window `4-Hz D2O lock set on-resonance`.
8. Click **Start Calibration**.
9. Click **Confirm** in the popup prompt window if a **PFG probe** is in the magnet.

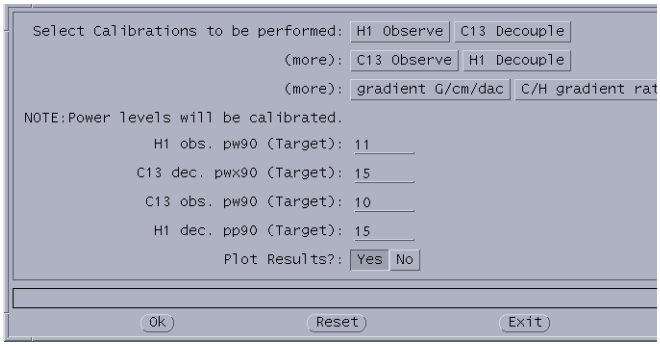
   At the end of the calibration routine, the calibrations are automatically incorporated into the probe file.

Calibrating Probe and System Files

1. Eject the sample from the magnet and insert the **1% ^{13}C-enriched methyl iodide, 1% trimethyl phosphite, and 0.2% Cr(AcAc) in chloroform-d** sample. Tune the probe if needed.
2. Click **Select Calibration** in the Probe window.
3. Set **AutoLOCK** and **AutoSHIM** to **YES**.
4. Right-click the **Experiment** drop-down menu and select **Calibrate H, C, Ind.Det.Grad (CH3I)**.



5. Click **Ok**.
6. Click **Exit**.
7. Click **Confirm** to confirm that the **correct** sample is in the magnet.
8. Click **Start Calibration**.
9. Select the following:



   H1 Observe
   C13 Decouple
   C13 Observe
   H1 Decouple

   If the probe is equipped with gradients, also select:

   gradient G/cm/dac
   C/H gradient ratio

   These are typical calibration for autoswitchable, indirect detection, and triple resonance probes.

10. Enter target values for **^{1}H obs pw90, ^{13}C obs pw90, ^{1}H dec pp90,** and **^{13}C dec pwx90** calibrations. The values are usually the pulse specifications for the probe.
11. For **Plot Results?**, select **Yes**.
12. Click **Ok**.
13. Click **Exit**.

At the end of the calibration routine, the power and pulse width values are automatically incorporated into the probe file.

2.2 Automated Data Acquisition

The automated data acquisition procedures outline several 1D and 2D experiments using the ethyl-indanone sample.

These experiments demonstrate the capabilities of the spectrometer, the correct calibration of the instrument, and validate the correct functioning of the instrument. These experiments do not use the sample changer. If a sample changer is present, set **`traymax=0`**.

The following is part of the ATP and training session:

- Set up and use Walkup interface.
- Use the application type selection drop down menu to select the application type, Std1D, Hetero 2D, and Homo 2D.
- Set up 1D, 2D, gradient (if appropriate hardware is installed) and non gradient protocols by selecting from the list of protocols the experimental protocols for proton and carbon 1D, homonuclear 2D, and heteronuclear 2D experiments.
- Create, run, save data, and plot the results obtained a composite protocol.

Use the **VnmrJ Walkup** interface for these experiments, it is a required part of the ATP. These experiments can be run using either the **Walkup Account Owner** or **Walkup Operator** interface.

Refer to the *VnmrJ Software Installation and Administration* manual for instructions on setting the user interface (both the walkup administrator and walkup operator) and the *Walkup VnmrJ* manual for working with the VnmrJ Walkup interface.

Sample for Automated Data Acquisition

Sample	*Sample Size (mm)*	*Sample Part Number*
2% 2-ethyl-1-indanone in chloroform-*d*	5	01-901855-03

Protocols for Automated Data Acquisition

The following protocols will be combined into a single composite protocol. The composite protocol is submitted to the study queue and the 1D and 2D experiments specified by the protocol are run on the study sample. This demonstrates the ease with which a study protocol can be created and run on a given sample.

System (console and probe) configuration determines the experiment selection as follows:

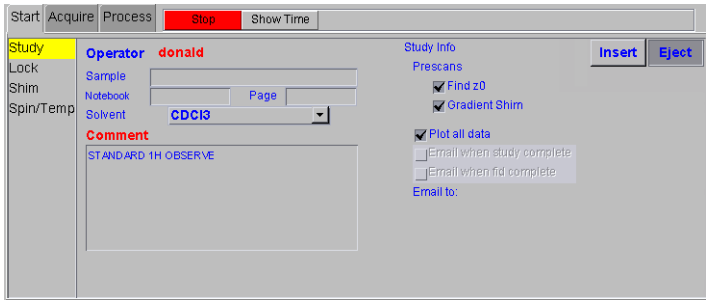
Protocol	*Gradient Systems*	*Nongradient Systems*
1 D Protocols:		
Acquisition of a proton spectrum	❑	❑
Acquisition of a proton decoupled carbon observe spectrum	❑	❑
Dept (distortionless enhancement by polarization transfer)	❑	❑
Apt (attached proton test)	❑	❑
Non-gradient 2D Protocols:		
Tocsy (total correlation spectroscopy)	❑	❑
Noesy (nuclear overhauser spectroscopy)	❑	❑
Gradient 2D Protocols (requires PFG option and gradient probe)		
Gcosy (gradient correlation spectroscopy)	❑	
Ghsqc (gradient heteronuclear single quantum correlation	❑	
Ghmbc (gradient heteronuclear multiple bond correlation)	❑	
Non-gradient Protocols		
Cosy (correlation spectroscopy)		❑

Login to Walkup Interface

1. Log on using the UNIX VnmrJ **Walkup Account Owner**.
2. Select the Walkup account owner from the Operator menu, enter the password, and click OK.
3. Select **Utilities** -> **New automation run**.
4. If a sample changer is present, set **`traymax=0`**.
5. Make sure the printer/plotter is set up, `pfgon` is set properly, and shim map that was made in 2.1 “Calibrating a Probe” page 15 has been copied into `/vnmr/shimmaps`.

Setting Up the Study and Lock Solvent

1. **Insert** the **indanone** sample (01-901855-03).
2. Click the **Start** tab.



3. Select the **Study** page.
4. Enter sample information in the **Comment** field:

`2-ethyl-1-indanone`

5. Enter information (Optional) in the **Notebook**, **Page**, and **Sample** fields**.**
6. Select **CDCl3** from the **Solvent** drop down menu.
7. Place a check in box to enable **Plot all data** option.
8. Make sure the **Find Z0** and **Gradient shim** boxes are **checked**.
9. Do not place a check in check boxes for:

   Email when study complete –or– Email when fid complete.
   If a check appears in either box, click the box to remove the check.

Building a Composite Protocol

1D Protocols

1. Click the **Std 1D** tab, and click the following experimental protocols in this order:
   a. **Proton**
   b. **Carbon**
   c. **Dept**
   d. **Apt**

2D Nongradient Protocols

1. Click the **Homo 2D** tab, and click the following experimental protocols in this order:
   a. **Tocsy**
   b. **Noesy**
   c. Cosy (click only if the system does not have gradients)
2. Continue with "2D Gradient Protocols," page 21 if the system has gradients
   –or–
   go to "Customizing the Parameters and Starting Data Acquisition," page 22 if the system does not have gradients.

2D Gradient Protocols

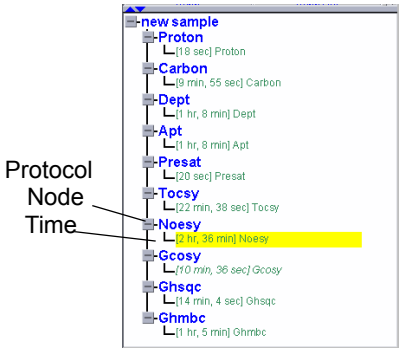
1. From the Homo 2D tab, click **Gcosy**.
2. Click the **Hetero 2D** tab, and click the following experimental protocols in this order:
   a. **Ghsqc**
   b. **Ghmbc**

Customizing the Parameters and Starting Data Acquisition

The study queue contains the protocols in the order each one was selected and should look similar to the study queue shown below.

- "Customize the Noesy Parameters," page 22
- "Customize the Carbon Parameters," page 22

Customize the Noesy Parameters



Study Queue

Customized NOESY Parameter

1. Click the **[+]** for the **Noesy** protocol if the protocol time is not visible.
2. Double-click the protocol time to retrieve the parameters.
3. Click the **Acquire** tab, select the **Defaults** page (if more than one page is available). The parameter panel and the pulse sequence are displayed.
4. Modify the acquisition parameter as follows for both Gradient and Non-Gradient systems or non-Gradient Probe.
5. Change the following acquisition parameters:
    a. Click the **Scans per increment** drop down menu and select **4**.
    b. Click the **Mixing time [ms]** drop down menu and select **1000.**

Customize the Carbon Parameters

1. Click the **[+]** for the **Carbon** protocol if the protocol time is not visible.
2. Double-click the protocol time to retrieve the parameters.
3. Click the **Acquire** tab and select the **Defaults** page (if more than one page is available).
4. Select **1000** from the **Number of Scans** drop down menu.
5. *Uncheck* **Test for S/N at every block size**.

Acquiring Data Using a Composite Protocol

1. Click the **Submit** button.
2. All the protocols are locked automatically.
3. Acquisition starts.

Continue with 2.3 "Magnet Drift Test" page 23.

2.3 Magnet Drift Test

The magnet drift test is an overnight test.

Samples for Magnet Drift Test

Use the sample that provides a signal with good signal-to-noise ratio, in most cases the 1% H_2O / 99% D_2O samples will a good signal, see Table 2.

Table 2. Sample for Magnet Drift Test

Sample	*Sample Tube (mm)*	*Sample Part Number*
Doped 4-Hz H_2O/D_2O (0.1 mg/ml $GdCl_3$ in 1% H_2O in D_2O)	5	01-901855-01
Doped 2-Hz H_2O/D_2O (0.1 mg/ml $GdCl_3$ in 2% H_2O in D_2O)	5	01-901855-02
autotest sample; 0.1% ^{13}C enriched methanol in 1% H_2O/99% D_2O	5	01-968120-68

Probe and Hardware Requirements

A 5-mm probe capable of ^{1}H direct observe is recommended.

Set up

1. Click the **Locator Statements** menu ( ) and select **System Param Files** from under the **Sort NMR Parameter Files** category.
2. Click the title above the right most column and select **Directory** from the lists that is displayed.
3. Select **`shmd2o`** and drag it to the VnmrJ graphics canvas.
4. Insert the **4-Hz H_2O/D_2O** sample.
5. Tune the probe.
6. Establish lock and adjust the field homogeneity.

Test Procedure

1. Select the **Future Actions** page.
2. The only option active on this page is **`wft`** in the **When Experiment Finishes** field. All other options should be disabled or the field blank.
3. Click the **Acquire** button and obtain a normal spectrum and shim the HDO signal to 4 to 5 Hz linewidth at 50%.
4. Click the **Start** tab and select the **Lock** page.
    a. Adjust **Z0** to be on resonance.

b. Adjust **Power**, **Gain**, and **Phase** for a steady lock level of about 80.

5. Select the **Spin/Temp** page and do the following:
   a. Click the **Ignore spinner error** radio button
   b. Click the **Ignore temperature error** button
   c. Enter **0** in the spin spinner speed field and click the **Spin Off** button.
6. Click the **Acquire** tab and select the **Acquisition** page.
7. Enter **1** in the **Scans Requested** field
8. Click the **Arrays** button and fill in the array form as follows:
   a. Enter **`d1`** in the cell under **Param Name**.

      The row will be highlighted and the cell descriptions and values will be filled in for the remaining cells. If this did not happen you did not press return after entering `d1` in the cell under Param Name.
   b. Enter **11** in the **Array Size** field.
   c. Enter **3600** in the **First Value** field.
   d. Enter **0** in the **Increment** field.
   e. Highlight the **first value** for the first position and enter **60**.
   f. Click **Close**.
9. Click the **Acquire** button.

   The test takes approximately 10 to 11 hours to finish. At the conclusion of the experiment the data are automatically processed and displayed.

   The data can be processed manually if desired:
   a. Click the **Process** tab and select the **Process** page
   b. Click **Transform All** then **Autophase Zero** buttons.
   c. Select the **Display** page.
   d. Set **Display Mode** to **Phased**.
   e. Set **Axis** to **Hertz**.
   f. Set **Amplitude scaling** to **Absolute**.
   g. Click **Full** button **Screen Position**.
   h. Click **Display Array** buttons **Vertical** and **Label**.
10. Compare the frequency shift of the HDO peak of the arrayed spectra to the frequency of the first spectrum in the array.
11. Write the results on the form in “Console and Magnet Test Results,” page 69.

2.4 Optional Tests

The following optional tests will be run if they are specified in contract and the hardware is present.

Variable Temperature Operation (Optional Hardware)

This demonstration shows that the basic variable temperature (VT) unit and probe changes to the desired temperature and displays on the VT controller. If the system is equipped with a VT unit, read through the VT operation instructions before this demonstration.

Dry nitrogen is required as the VT gas if the requested temperature is over 100° C or below 10° C. Otherwise, air can be used. Dry nitrogen gas is recommended for cooling the bearing, spinner, and decoupler. This prevents moisture condensation in the probe and spinner housing.

CAUTION: **The use of air as the VT gas for temperatures above 100° C is not recommended. Such use destructively oxidizes the heater element and the thermocouple.**

Demonstration Limitations

If dry nitrogen gas and liquid nitrogen are unavailable at the time of installation, the range of VT demonstration is limited to temperatures between 30°C and 100°C.

Sample

No sample is used.

Probe and Hardware Requirements

Any VT probe is used.

Procedure

1. Click the **Utilities** menu and select **Systems Settings**.
2. Click the **System config** button and verify the VT Controller is set to Present.
3. Set N2 gas flow to 9.5 to 10.0 LPM (for temperatures below –100° C, increase N2 flow to 12 LPM).
4. Click the **Start** tab and select the **Spin/Temperature** page.
   a. Enter a value in the temperature field (or use the slider bar to set a value).
      The heat exchanger must be in place for values below room temperature.
   b. Click the **Regulate Temp** button.
      Maintain the temperature for 5 minutes.
5. Operate the VT unit within the specifications of the probe. Test the temperature at set points that correspond to the following:

- Maximum, minimum, and midpoint of the allowed temperature: 95, 80, 60 if air is used; 120, 30, 20 if dry nitrogen is used; or 120, –100, 40 if a heat exchanger is used.
- Ambient temperature.

The software limits the ramp rate to 12°C per minute up or down. Wait for the temperature to equilibrate.

Temperature Accuracy for VT Systems (Optional Test)

The optional tests in this section check temperature accuracy calibrations for high and low temperatures using ethylene glycol and or methanol, respectively.

Table 3 lists the samples for low-temperature and high-temperature tests.

Table 3. Samples for Optional VT Accuracy Test

Sample	*Temperature Range (°C)*	*Sample Tube (mm)*	*Sample Part Number*
100% methanol (reagent grade)	–50 to +25 (Low)	5	00-968120-80
100% ethylene glycol (reagent grade)	+25 to +100 (High)	5	00-968120-79

Probe and Hardware Requirements

The variable temperature accessory and a VT probe are required. Run VT tests with a 5-mm probe capable of ^{1}H direct observe from –150°C to +200°C. For probes that have a more limited temperature range (particularly PFG probes), run the test at two or three temperatures that fall within the VT range of the probe. These tests can also be run using the ^{1}H decoupling coil of the 5-mm broadband probe as ^{1}H direct observe.

Set up

1. Click the **Locator Statements** menu ( ) and select **System Param Files** from under the **Sort NMR Parameter Files** category.
2. Click the title above the right most column and select **Directory** from the lists that is displayed.
3. Select **`shmd2o`** and drag it to the VnmrJ graphics area.
4. Insert the **doped 4-Hz H_2O/D_2O** sample.
5. Make sure that the VT gas flow and cooling air flow levels are between 9.5 to 10 LPM and gas flow to the probe is not restricted in any way.
6. Set up the parameters. Acquire a normal spectrum and shim the water signal to about 3 to 4 Hz linewidth at 50%.
7. Click the **Start** tab and select the **Study** page.
   a. Eject the doped 4-Hz H_2O/D_2O sample
   b. Insert the appropriate sample:

      High Temperature— 100% ethylene glycol (00-968120-79).

      Low Temperature— 100% methanol (00-968120-80).
   c. Click the drop down menu next to **Autolock** and select **Unlocked**.

8. Select the **Lock** page and click the lock **OFF** button.
9. Click the **Acquire** tab
   a. Select the **Acquisition** page.
   b. Set the following parameters: **Observe Pulse** to **2**, **Receiver Gain** to **5** (or some value that doesn't overload the receiver), **Spectral width** to **10000**, **Acquisition time** to **2**, and **Scans Requested** to **1** by entering these values in the fields next to the parameters.
   c. Select the **Future Actions** page.
   d. Remove any entries in the ***If an Error Occurs*** field.

   The test is run unlocked, because the sample has no deuterated solvent to lock on.
10. Click the **Start** tab and click **Setup Hardware**. Check the probe tuning for the ethylene glycol sample.
11. Click the **Acquire** button to acquire the spectrum.
12. Place the single cursor between the two peaks.
13. Click the **Process** tab and select the **Cursor/Integration** page.
14. Click the **Move transmitter** button.
15. Make sure the VT gas flow and cooling air flow levels are between **9.5 and 10 LPM**.

Data Acquisition

1. Acquire a spectrum, record the temperature, and record the chemical shift.
   a. Click the **Acquire** button to acquire another spectrum.
   b. Select the  icon from the graphics control bar.
   c. Position the right and left cursors on the right and left peaks.
2. Pull down the command line and do either of the following:
   - **High Temperature**— Enter `tempcal('glycol')`.
   - **Low Temperature**—Enter `tempcal('methanol')`
3. Record the temperature reading on the VT controller.

   Temperature is displayed on the face of the VT controller, remote status unit (if ordered, and on the hardware bar.
4. Record the chemical shift frequencies of the two peaks.
5. Follow the procedure for either the high or low temperature test:
   - "High-Temperature Calibrations Test," page 27
   - "Low-Temperature Calibrations Test," page 28

High-Temperature Calibrations Test

CAUTION: **Extreme temperatures can damage the probe. The high and low temperatues must be within the specified range of the probe.**

1. Follow the procedure in "Set up," page 26.
2. Acquire a spectrum at room temperature, follow the procedure in "Data Acquisition," page 27.

3. Click the **Start** tab and select the **Spin/Temperature** page.
   a. Enter `50` in the temperature field (or use the slider bar to set a value).
   b. Click the **Regulate Temp** button.
      Wait a minimum of 10 minutes for the temperature reach regulation.
4. Repeat the procedure in "Data Acquisition," page 27.
5. Click the **Start** tab and select the **Spin/Temperature** page.
   a. Enter `100` in the temperature field (or use the slider bar to set a value).
   b. Click the **Regulate Temp** button.
      Wait a minimum of 10 minutes for the temperature reach regulation.
6. Repeat the procedure in "Data Acquisition," page 27.

Low-Temperature Calibrations Test

CAUTION: **For low-temperature calibrations, fill the VT dewar with liquid nitrogen. If a chemical mixture is used instead of liquid nitrogen for low-temperature calibrations, choose the chemical slurry carefully. A mixture of crushed dry ice and acetone is not recommended, because it will dissolve the polystyrene VT dewar.**

1. Follow the procedure in "Set up," page 26.
2. Acquire a spectrum at room temperature, follow the procedure in "Data Acquisition," page 27.
3. Click the **Start** tab and select the **Spin/Temperature** page.
   a. Enter `-20` in the temperature field (or use the slider bar to set a value).
   b. Click the **Regulate Temp** button.
      Wait a minimum of 10 minutes for the temperature reach regulation.
4. Repeat the procedure in "Data Acquisition," page 27.
5. Click the **Start** tab and select the **Spin/Temperature** page.
   a. Enter `-80` in the temperature field (or use the slider bar to set a value).
   b. Click the **Regulate Temp** button.
      Wait a minimum of 10 minutes for the temperature reach regulation.
6. Repeat the procedure in "Data Acquisition," page 27.
7. After finishing the low-temperature test:
   a. Click the **Start** tab and select the **Spin/Temperature** page.
   b. Click the **Temp Off** button
   c. Click the **Reset VT** button
   d. Keep the dry nitrogen gas flowing to the probe and upper barrel
   e. Remove the polystyrene VT dewar containing liquid nitrogen.
   f. Continue the flow of dry nitrogen gas to the probe to prevent condensation inside the probe. Allow the dry nitrogen gas to flow through the probe and upper barrel for at least 15 minutes while the probe warms up to room temperature.

Data Analysis

Plot a graph of the VT controller reading (horizontal axis) as compared with the calculated VT reading from the chemical-shift differences between the two peaks (vertical axis). Draw a straight line through the points.

Stability Calibration for High-Stability VT (Optional Test)

This optional test is for high-stability VT units only (Part No. 00-992953-00). The test demonstrates that the VT unit can hold the temperature with ± 0.1°C. The test requires preconditioning of the laboratory air and restricts the room temperature fluctuations.

Table 4. Samples for Optional High-Stability VT Test

Test Sample	*Nucleus*	*Sample Tube (mm)*	
10-mM DSS in D_2O (sample volume of 0.6 ml in a 5-mm NMR tube))	^1H	5	Customer supplied

DSS= 3-(trimethylsilyl)-1-propanesulfonic acid. The customer must make this sample using DSS and deuterium oxide (99.8 or 99.9 atom%D). Upon request, Varian can make this sample if DSS is not available at the customer site.

Probe and Hardware Requirements

High-stability variable temperature accessory and a 5-mm probe capable of ^1H direct observe are required.

Test Procedure

1. Click the **Locator Statements** menu ( ) and select **System Param Files** from under the **Sort NMR Parameter Files** category.
2. Click the title above the right most column and select **Directory** from the lists that is displayed.
3. Select **`shmd2o`** and drag it to the VnmrJ graphics canvas.
4. Insert the **doped 4-Hz H_2O/D_2O** sample (01-901855-01).
5. Click the **Acquire** tab
   a. Select the **Acquisition** page.
   b. Set the following parameters: **Observe Pulse** to **90** degree pulse width for the probe, **gain** to a value that doesn't overload the receiver, **Spectral width** to **10000**, **Acquisition time** to **10**, and **Scans Requested** to **1** by entering these values in the fields next to the parameters.
   c. Select the **Future Actions** page.
   d. Remove any entries in the ***If an Error Occurs*** field.
6. Click the **Start** tab and select the **Spin/Temperature** page.
   a. Enter **`40`** in the temperature field (or use the slider bar to set a value).
   b. Click the **Regulate Temp** button.
   Wait a minimum of 10 minutes for the temperature reach regulation.

c. Set spin to 0.

Allow the VT controller to regulate to 40° C, which should be about 10° higher than the room temperature. Check that the probe is tuned.

7. Click the **Acquire** button to acquire the spectrum.
8. Place the single cursor between the DSS signal (right-most peak).
9. Click the **Process** tab and select the **Cursor/Integration** page.
10. Click the **Move transmitter** button.
11. Click the **Acquire** tab
    a. Select the **Acquisition** page.
    b. Set **Spectral width** to **1000**.
12. Click the **Acquire** button to acquire the spectrum.

    Shim the DSS signal to about 0.6 Hz or less linewidth at 50%. The sample of DSS in D_2O should equilibrate at 40° C for at least 2 hours before the next step.
13. Click the **Arrays** button and fill in the array form as follows:
    a. Enter **`d1`** in the cell under **Param Name**.

       The row is highlighted and the cell descriptions and values are filled in for the remaining cells.
    b. Enter **73** in the **Array Size** field.
    c. Enter **600** in the **First Value** field.
    d. Enter **0** in the **Increment** field.
    e. Highlight the **first value** for the first position and enter **0**.
    f. Click **Close**.

    This sets up an array of `d1` values with the first spectrum, collected at time 0 minutes, and subsequent spectra, collected at 10 minute intervals for up to 12 hours.
14. Click **Acquire**. The test takes about 12 hours to complete.
15. After the data acquisition is completed, the data is processed and the spectra are displayed as a stacked display.
16. The data can be processed manually as follows:
    a. Click the **Process** tab and select the **Process** page
    b. Click **Transform All** then **Autophase Zero** buttons.
    c. Select the **Display** page.
    d. Set **Display Mode** to **Phased**.
    e. Set **Axis** to **Hertz**.
    f. Set **Amplitude scaling** to **Absolute**.
    g. Click **Position** button **Full**.
    h. Click **Display Array** buttons **Vertical** and **Label**.
17. Measure the difference between the left-most peak and the right-most peak in Hz.

Chapter 3. Consoles and Magnets Specifications

This section contains the following specifications:

3.1 Magnet Drift

Table 5 lists the drift specifications for magnets. Specifications for nominal field decay rate are less than or equal to the values listed in the table.

Table 5. Magnet Drift Specifications

System (MHz/mm)	*Field Strength (T)*	*Nominal Field Decay Rate (Hz/hr)*
200/54	4.70	2
300/54	7.05	3
400/54	9.40	8

3.2 Variable Temperature Operation

For basic variable temperature (VT) accessories (Varian Part No. 00-992957-00), demonstrate that the VT unit and probe go to the desired temperature as registered on the window of the VT controller. If the system is equipped with a VT unit, the system user should read through the VT operation instructions before the demonstration.

Dry nitrogen is required as the VT gas if the requested temperature is over 100°C or below 10°C. Otherwise, air can be used. For temperatures below –40°C, dry nitrogen gas is recommended for cooling the bearing, spinner, and decoupler. This prevents moisture condensation in the probe and spinner housing.

CAUTION: **The use of air as the VT gas for temperatures above 100°C is not recommended. Such use destructively oxidizes the heater element and the thermocouple.**

CAUTION: **Extreme temperatures can damage the probe. The high and low temperature must be within the specified range of the probe.**

Demonstration Limitations

If dry nitrogen gas and liquid nitrogen are not available at the time of installation, the range of VT demonstration is limited to temperatures between 30°C and 100°C.

Basic Specifications

The specifications for variable temperature ranges are listed with each probe.

3.3 Temperature Accuracy for VT Accessories

The temperature reading displayed on the VT unit display panel should be within ±1° C of the actual temperature reading, as measured from the chemical shift.

3.4 Stability Calibration for High-Stability VT Accessory

The high-stability VT accessory holds the set temperature to within ±0.1°C. (±0.1°C = 0.001 ppm or in field dependent terms:, ±0.2 Hz at 200 MHz, ±0.3 Hz at 300 MHz, and ±0.4 Hz at 400 MHz)

Chapter 4. Customer Training

Sections in this chapter:

This chapter describes training provided by the installer. This training is intended as a general overview of the instrument, basic maintenance requirements, software features, data acquisition and storage, file maintenance, and other routine tasks. Comprehensive training classes are offered at various Varian, Inc. Applications Laboratories around the world. Call your sales representative or contact the Varian, Inc. NMR systems office nearest you for class offerings, schedules and cost.

4.1 Where to Look for Answers

Knowing where to look is the first step to answering a question. VnmrJ provides two libraries of information:

- “VnmrJ Online Help” on this page
- “Online Manuals” on this page

VnmrJ Online Help

VnmrJ provides online help – click **Help** on the VnmrJ main menu and select **VnmrJ Help**. An internet browser is started. Navigate the help system using the available browser tools.

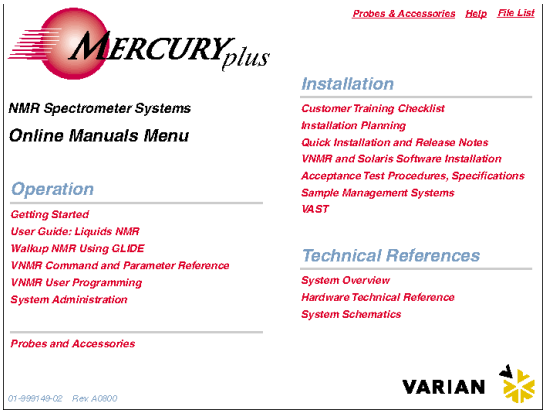


Figure 2. Example Online Manual Menu

Online Manuals

The complete library of manuals related to the system, software, and available accessories is accessible as PDF files. Figure 2 shows an example menu for the online manuals. After the installation has been finished you will find many of your routine questions answered in the following manuals.

- *VnmrJ Walkup NMR* – Provides:
  - — information about the walkup NMR interfaces available in VnmrJ.
  - — step-by-step instruction on setting up experiments
  - — instructions for automated sample changer operation.
- *VnmrJ Liquids NMR*– Provides:
  - — an overview of VnmrJ and instrument operations.
  - — instructions for locking, shimming, and probe tuning.
  - — explanations of data acquisition parameters and digital signal processing
  - — data processing, display, and plotting.
  - — instructions for setting up all the standard experiments provided with VnmrJ.

    Not every experiment listed can be run on every system. The type and configuration of the instrument will determine which experiments can be run. This is the manual to go to when you have very specialized experimental requirements and will most likely be running the experiments from the command line

Installations

- *Installation Planning Guide* – Provides:
  - —site planning information and site requirements.
- *Acceptance Test Procedures* – Provides:
  - — installation test procedures and specifications for spectrometer and magnet.

    Included in this manual is the information used by the installation engineer for the introductory training at the end of the installation. This manual does not cover the probe test procedures, but refers to them.
- *VnmrJ Installation and Administration*– Provides:
  - — instructions for the installation VnmrJ software.
  - — instructions for setting up various plotters and printers.
  - — instructions for various VnmrJ related administration tasks.
- *Solaris Installation and Administration*– Provides:
  - — instructions for the installation Solaris software.
  - — instructions for various Solaris related administration tasks.
- *Linux Software Installation*– Provides:
  - — instructions for the installation Linux software.
- Probe installation, testing, and specifications manuals – Each type of probe has its own installation, test, and specifications manual. In these manuals are the detailed instructions for installing, tuning, testing the probe.
- Accessory manuals – Each accessory is covered in an independent manual, which contains installation, testing, and sometimes operation instructions.

Technical References

- S*ystem Schematics* – Provides:
  - — schematic and technical drawings.
- *Technical Reference* – Provides:
  - — technical details of the spectrometer systems and electronics.
- *System Description* – Provides:
  - — an overview of the spectrometer system and hardware.
- *User Programming* – Provides:
  - — details about the VnmrJ macro programming language Magical II.
  - — instructions for writing custom macros and editing existing macro.
- *Command and Parameter Reference* – Provides:
  - — Alphabetical reference for commands, parameters, and macros.

4.2 VnmrJ Directory Structure

The *VnmrJ Installation and Administration* manual contains detailed information about the VnmrJ directory structure.

The VnmrJ directory and file structure is set up with both global files and directories and user accounts with user level files and directories. Directories and permission levels for UNIX logins other than root are as follows:

Directory	*Description*	*Read, Write, and Execute Permission*
/export/home/vnmr		
	System or global directories and files	VnmrJ administrator (typically vnmr1)
/vnmr		
	Symbolic link to: `/export/home/vnmr`	VnmrJ administrator (typically vnmr1)
/export/home/<user>		
	User directory and files	UNIX log in account owner
/export/home/<user>/vnmrsys		
	User VnmrJ directories and files	UNIX log in account owner

The VnmrJ administrator, rather than root, should make changes to VnmrJ files and directories. The global files and directories contain macros, pulse sequences, binary files, optional VnmrJ software, and other files that have a common usage to all users.

When a command or instruction is executed in VnmrJ by a user, VnmrJ first searches the ~/`vnmrsys/` directory for the command, pulse sequence, etc. and then searches the global files in `/vnmr`. When creating custom macros, pulse sequences, menus, etc. it is necessary to give these files different names from the similar files in `/vnmr`. If you want the customized macros, pulse sequences, etc. to be available to all users, place them in the appropriate directory in `/vnmr`.

4.3 Initial System Settings

System settings are used to enable or disable installed hardware and define the events that will occur upon completion of data acquisition. System settings do not replace the system configuration. Keep the defaults except as described below:

1. Click the **Utilities** menu and select **System Settings**.
2. Click the **System** tab.
3. Enable the Z gradient. Select **on** from the drop down menu next to the **Z** to the right of **Gradient amplifier**.
4. Set **Type of digital signal processing** to **Realtime**.
5. Do not enable **Frequency-shifted quadrature detection** (no check in the box).
6. Set **Hardware Z1 shimming** to **None**.
7. Do not enable **Probe protection** or **Solids VT System** (no check in the boxes).
8. Set **VT cutoff (0-50)** to **25**.
9. Enable **Process data after acquisition** and **Autosave data after acquisition** (check in the box).
10. Click the **Display/Plot** tab.
11. Enable **Process data on drag–and–drop** (check in the box).
12. Set **Spectrum updating during phasing (0-100)** to **100**.
13. Do not enable **Display only matching items in locator** (no check in the box).
14. Enable **Show current operator studies only** (check in the box).

4.4 Set Up Experimental and Walkup Users

Creating a new user account is described in the *VnmrJ Installation and Administration* manual. During the VnmrJ installation process the VnmrJ administrator (typically vnmr1) is created and the VnmrJ administration interface is run from this user account.

Each user account is assigned to an interface type: Experimental, Walkup, or Imaging. During the training process, both the Experimental and Walkup interfaces are used.

- The calibration procedures are run by the VnmrJ administrator (typically vnmr1) using the Experimental interface.
- The automated data acquisition procedures are run by either the walkup account owner or by an operator that does not have UNIX system privileges but has NMR operating privileges by way of the Walkup Operator interface.

1. Login as the VnmrJ Administrator (typically vnmr1).
2. Open a terminal window.
3. Enter **`vnmrj adm`** — the VnmrJ Administration interface starts.
4. Set the VnmrJ Administrator account to use the Experimental interface as follows:
   a. Click on the VnmrJ administrator login in the user panel.
   b. Click on the radio button next to **Experimental** in the user profile panel if this button is not already selected.

c. Click on **Save User** in the menu bar.

5. Set a user for the Walkup interface as follows:
   a. Click on the user's login in the users panel.
   b. Click on the radio button next to **Walkup** in the user profile panel if this button is not already selected.
   c. Click on **Save User** in the menu bar.
6. Exit the VnmrJ Administration interface.
   a. Click on **Management** in the menu bar.
   b. Click on **Exit** — the interface closes.

4.5 Basic Spectrometer Operation

This section contains exercises that are designed to acquaint you with basic spectrometer operations using the walkup and experimental interfaces in VnmrJ. These exercises should take no longer that 2 hours to complete.

Before proceeding, the system must be set up and calibrated as described in Chapter 2, "Console and Magnet Test Procedures," of this manual. The first 3 exercises will use the Walkup mode, so be sure you are logged in as the walkup user for these.

Sample for Exercises

Sample	*Sample Size (mm)*	*Sample Part Number*
2% 2-ethyl-1-indanone in chloroform-*d*	5	01-901855-03

Disable the Sample Changer

It is easier to run the exercises without using a sample changer, so if your system has a sample changer, disable it. You can reenable it after the finishing the exercises.

- To *disable the sample changer*, enter **`traymax=0`**. You will need the command line, which is not normally displayed. To display it, simply click on the small dot at the top-center of the graphics area and drag it down.)
- To *enable the sample changer* (after finishing the exercises), reset `traymax` to its proper value (or exit from and restart VnmrJ to automatically resets traymax).

Walkup Interface

Refer the *VnmrJ Walkup* manual and review the description of the Walkup interface.

Prepare and Insert the Sample

1. Put the NMR sample tube into the turbine (sometimes called the spinner). Use the sample depth gauge to center the sample volume. Refer to section *2.3 "Preparing the Sample,"* of the *VnmrJ Liquids NMR User Guide* manual.
2. Click the **Start** tab and select the **Study** page. Click the **Eject** button and then place the sample/turbine into the top of the upper barrel. Click the **Insert** button to lower the sample/turbine into the probe.

Exercise 1: 1D Proton Using the Study Queue

The purpose of this first exercise is to acquaint you with one of the standard ways to set up routine experiments, and with the things that normally have to be done with most experiments.

Before you begin:

- you must be logged in as a walkup user
- the sample must be prepared and inserted as described above.

Set Up and Acquire 1D Proton

1. Select the **Std 1D** tab, and click *once* on **Proton**.

   You will see a new sample entry in the Study queue area (the lower-left portion of the window).
2. Double-click **[1 min] Proton** to load the experimental parameters.
3. Select the **Study** page and set **Solvent** to **CDCl3**.
4. Enter whatever you like (or nothing at all) for Sample, Notebook, Page and Comment.
5. Under **Prescans** select **Find z0** and **Gradient Shim**. You can choose the **Plot all data** option, or not, as you wish.

   The first study is now ready for acquisition.
6. Click the **Submit** button to start the acquisition. The Submit button is under the Study Queue.

Observations

The first thing you should see happening (after some overhead, during which the study status changes from Queued to Active) is a 1-scan spectrum, the Find z0 scan, which sets z0 for (later) autolocking.

After the Find z0 scan is completed gradient shimming is set up and started; during both the Find z0 scan and gradient shimming the Proton node shown in the Study queue area will be highlighted in yellow. The shimming will probably take 2 or 3 iterations, and should be completed in 3-4 minutes, after which the actual experiment starts.

The highlight color changes from yellow to turquoise, showing the currently active experiment. When the experiment is complete the data is automatically loaded into the

current workspace, where it can be viewed. (For a description of the interactive display see the *VnmrJ Liquids NMR* manual.)

Summary

Locking, shimming, acquisition, processing, plotting, and saving the data was done automatically, requiring only what you selected in the Study parameter panel. The flow of events was:

- select the experiment
- select experimental options
- acquire data (including Find z0, gradient shimming, and the actual acquisition)
- process, plot, and save data

In the coming exercises we will learn more about doing these things by hand.

Exercise 2: 1D Proton and COSY Using the Study Queue

The purpose of this exercise is to further explore the Study Queue, introducing a simple 2D experiment to see how a series of experiments can be run on a single sample. You will also learn about using multiple workspaces to view several sets of data.

Setup and Acquire the COSY

1. Select **New sample** from the **Study Options** menu.
2. Select the **Homo 2D** tab, and click once on **Gcosy** (just **Cosy**, if your system does not have gradients).

   Notice that the new sample entry in the Study Queue area shows both Proton and Gcosy nodes. Since the sample is already in the magnet there is no need to eject or insert it, and for that matter no need to lock and shim again (assuming this exercise is being done immediately after completing Exercise 1; if not, then you might need to do all these things). Therefore, we will need to customize this acquisition to turn off those choices (among other things).
3. Double click **[1 min] Proton** to initiate customization. The node is the actual entry showing the time (e.g. `[1 min] Proton`), not the bold Proton shown just above it.
4. Click the **Start** tab and select the **Study** page.
5. As before you can enter sample information and a comment; this time you want to make sure that both the **Find z0** and **Gradient Shim** are **not checked**.

   In two-dimensional (2D) experiments, using the entire (standard) H1 spectral window (-2 to 14 PPM) is usually efficient when running the 2D experiment. Therefore in addition to an initial wide-window 1D spectrum, we would like to acquire a second (1D) spectrum with the window reduced to just the relevant portion of the spectrum.
6. Click the **Acquire** tab, select the **Prescan** page, and select the **Minsw** option.

   It is also inefficient to acquire too many (or too few) increments in 2-dimensional experiments, so we would like to set the number explicitly. (It is not that the experiment would not be run well even without this step, but rather that the point is to learn what can be done, and how to do it.)
7. Double-click (or drag-and-drop) the **[4 min] Gcosy** node.

8. After the Gcosy pulse sequence is displayed, click the **Acquire** tab and select the Defaults page. From the **Number of increments** menu select **128**.

   Do not worry about the spectral width (shown as the standard full window); the value used in the actual experiment will be taken from the results of the Minsw prescan.

9. From the **Fourier number in F2&F1** menu, select **not used**.

10. Click **Show Time** to update the time estimate for this experiment.

11. Click **Submit** to acquire data.

Observations

The actual 1D proton acquisition is started immediately, using the standard (wide) spectral width. When it is complete,

- data is processed
- the spectral window adjusted to just the region of the spectrum containing signals
- and a second 1D proton spectrum is acquired, processed, plotted and saved.

Finally, the Gcosy (or Cosy) parameters are set up, and that spectrum acquired, processed plotted, and saved. On the plot, the high-resolution 1D proton spectrum should be plotted along the sides of the 2D (contour) plot.

You should also see that while the 1D spectra are being acquired it is the Proton node in the Study Queue that is highlighted (in turquoise), with the highlight on Gcosy during that experiment.

View the Proton Data

To view the Proton (strictly, the Proton_Minsw) data while the Gcosy data is being acquired, double-click the **Proton** experiment. The data is recalled, processed, and displayed.

Create a New Workspace

As in the first exercise, everything is done automatically. You are left with the COSY data in the current experiment. But what if you want to look at the 1D spectrum? It was saved after being acquired, but the data is no longer present in the current workspace (now being used for the COSY data).

You can reload the 1D data, but if you do that in the current workspace it will replace the COSY data, so you may want to use another workspace instead.

1. Select **Create a Workspace** under the **Utilities** menu (Utilities->Create a Workspace).

   A list of experiments appears in the locator.

2. Double-click **exp2** in the locator.

3. Click the **Home** button above the file browser.

   The easiest (but not the only) way to retrieve your data is from the Locator.

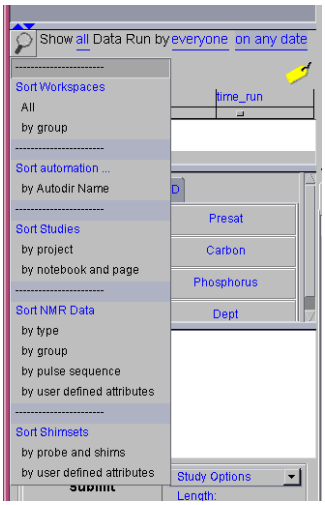
4. Click on the **Locator Statements** icon, and select Sort NMR Data.

   The search statement appears next to the Locator Statements icon,

   `Show all Data Run by everyone on any date`

5. Right-click everyone, and select me. Click the square underneath the time_run to sort the data by time. Drag your proton 1D experiment into the graphics window.

   It is automatically processed. Selecting Sort Workspaces from the Locator Statements will once again list exp1, exp2, etc in the locator. Switch between workspaces by double-clicking on the experiment.



Exercise 3: 1D Proton, HSQC, and More

The purpose of this exercise is to:

- introduce the Hetero 2D experiments in Walkup
- select multiple experiments
- remove experiments.

Queue Experiments

VnmrJ runs the experiments in the order they appear in the queue. Also, if 1D spectra are available, the spectra are plotted on the edge of the 2D plot. If no ^{13}C 1D spectrum is available, an F1 projection will be plotted instead.

1. Select the **Hetero 2D** tab.
2. Click once on the **Ghsqc** button (or Hsqc if your system does not have gradients). Notice, as before, a Proton 1D is also queued.
3. Click the **Ghmbc** button (or Hmbc if your system does not have gradients). The Ghmbc is added to the queue.
4. Select the **Std1D** tab. Click **Carbon** and drag it into the Study Queue between the Proton and Ghsqc. The Carbon experiment will be run before the HSQC.

Remove an Experiment

Removing experiments from the locator is just as easy.

1. Click the **Ghmbc** (the larger blue letters). A yellow box appears around the Ghmbc.
2. Drag it to the trash can.

Run the Hetero 2D Experiments

1. Double-click **[1 min] Proton**, and as before, select the **Start** tab then the **Study** page.
2. Enter sample and text information if desired. Also, if your sample is already locked and shimmed, deselect the Find z0 and gradient shimming boxes. Make sure the solvent is set to $CDCl_3$.

3. Select the **Acquire** tab and click on the **Prescan** button. Make sure **Proton** and **MinSW** are selected.
4. Double-click the **Carbon** node (`[10 min] Carbon`).
5. Select the **Acquire** tab.

   You can change the spectral width either by choosing one of the values from the pull down menu, or by specifying the upfield and downfield values. For example, change the –15 Upfield value to –5. Notice the spectral width [ppm] menu is now blank. You can also change the number of transients and relaxation delay. An option to test for signal-to-noise test at the completion of each block is also available. If this option is chosen, the acquisition will stop when the specified signal to noise is reached.
6. Double-click **Ghsqc**.
7. Select the **Acquire** tab.

   Here you can change the number of transients and the number of *t*1 increments. You can also turn off multiplicity edit from here.
8. Select the **Acquisition** page. Change the relaxation delay to 1 second.
9. Click on the **Submit** button.

Exercise 4: 1D Spectra Using the Experimental Interface

This exercise demonstrates using the Experimental interface. Before you begin, make sure of the following:

- a user account is set up to use the Experimental interface, as described in 4.4 “Set Up Experimental and Walkup Users” page 36.
- printers are set up and gradients are turned on for the user account.
- the name of the probe is the system probe.

Change to the Experimental Interface

1. If you are logged in as a Walkup user, exit VnmrJ. Select **Utilities** -> **Exit VnmrJ**.
2. Log out of Solaris by clicking **Exit** on the **CDE** menu bar.
3. After the Solaris login window appears, log in as the experimental user.
4. Start VnmrJ by entering **`vnmrj`** in a Terminal window.

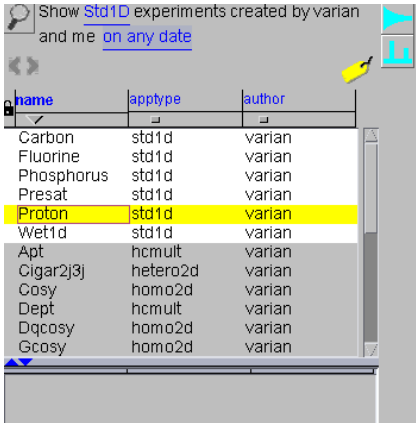
Setup and Acquire 1D Proton

As a first example, we will rerun the 1D proton spectrum, assuming in this case that the sample has just been put into the magnet and so needs to be locked and shimmed. You will be selecting the experiment from the locator.

1. Check to see that the upper level directory in the **File Browser** is **`/export/home`**. If not, click **Home** and then click the **up arrow** until only `/export/home` shows.

2. Click the **Locator Statement** menu and select **Sort Protocols**.

   A list of standard experiments should appear in the locator.



3. Double-click on the **Proton** experiment (or drag the icon from the Locator into VnmrJ graphics window).
4. Click the **Start** tab and select the **Standard** page.
5. Fill out sample information, select solvent, and add comments.
6. Click the **Find z0** button.

   The system acquires data and writes out a message with the z0 value.

7. Click on the **Gradient Shim** button.
8. After the system finishes gradient shimming, select the **Lock** page, and click the **Lock On** button.
9. Click the **Acquire** tab. Set the following as appropriate, although the default values are good for this example:
   - Number of Transients
   - Spectral Width
   - Relaxation Delay
10. Click on the **Acquire** button to start the acquisition.

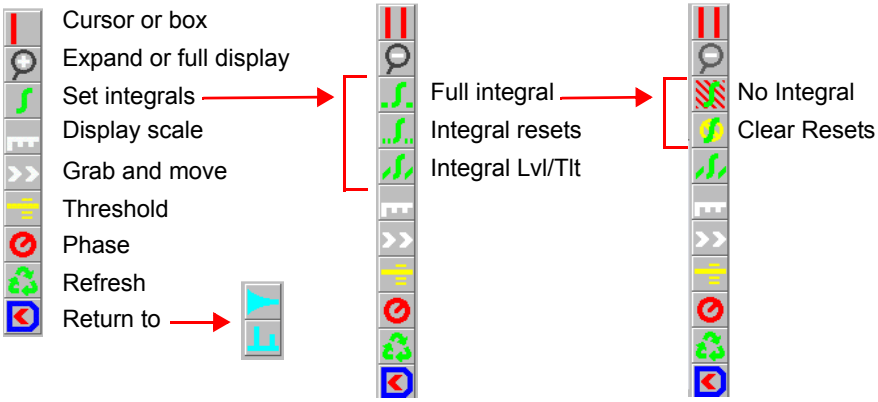
    The spectrum is displayed, but not saved. First we will save the data, and then demonstrate integration and custom plotting.

Save the Data

1. Click the **Acquire** tab and select the **Future Actions** page.
2. Click the **Save FID Now** button.

   The FID is be saved in your local data directory. Alternatively, you can select Automatic FID save before you start your experiment, and the FID will automatically be saved in your local data directory when the experiment is done.

Integration

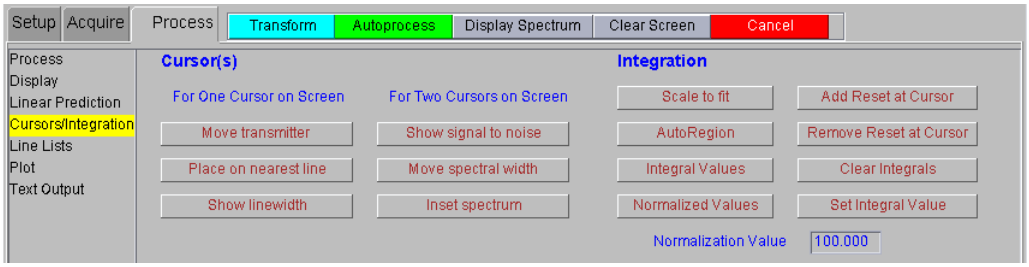


1. To reset the integral regions by hand, click the **Set integrals** icon in the graphics controls bar to the left of the spectrum.

   This will put you into partial integral mode, and it is from here you can reintegrate the spectrum by hand. You will now see three integral buttons. The middle button is the button used to select the regions to be integrated. You can click on the baseline or the integral itself to pick the regions.

2. With the ^{1}H 1D spectrum of the ethyl indanone displayed, select each peak as a region. You can expand the spectrum to pick regions if you wish.

3. After you are happy with the selected regions, click on the **Process** tab and then choose the **Cursors/Integration** page.



4. You can use this page to set the integral value for a region.For example:

   a. Click on the **methyl peak** (1 ppm) with the left mouse button so that the cursor is under the integral.

   b. Change the **Normalization Value** to **3.000**.

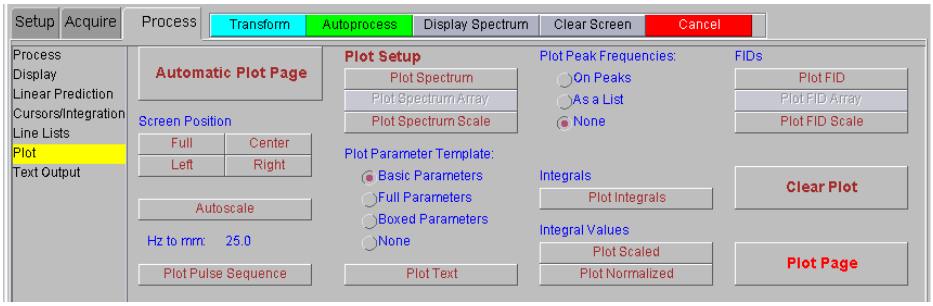
   c. Click the **Set Integral Value** button.

   The integral values will be displayed below the spectrum. If you wish to start over, the Clear Integrals buttons will remove all the resets.

Note that the integrals in the aromatic region will not correlate well because the aromatic protons have much longer *T*1s than the aliphatic protons).

Plotting

1. Click the **Process** tab and select the **Plot** page.



2. Click **Automatic Plot Page**.

   Refer to the *VnmrJ Liquids NMR* manual for information on customizing the plot.

Exercise 5: Running (Just) a COSY

Setup and Acquire the COSY

1. Redisplay the spectrum.
2. Click on the **Process** tab and select the **Process** page.
3. Click the **Transform all** button.
4. Place the cursors around the ^{1}H spectrum to select the desired spectral width.
5. Click the **Cursors/Integration** page and click the **Move spectral width** button.
6. From the **Experiments** menu, select:
   Homonuclear Correlation Experiments-> **Gcosy** (or Cosy if you do not have gradients).
7. Click the **Acquire** tab and select the **Defaults** page. Change the Number of Scans and Relaxation Delay as appropriate, or keep the defaults, which are fine for this example.
8. Click the **Acquire** button to start the acquisition.
9. After the acquisition has finished, click the **Process** tab and select the **Plot** page. Click the **Automatic Plot Page** button.

Observations

How does this plot compare to that produced using Study Queue (Exercise 1)?

Notice that the spectra plotted along the sides of the contour plot are NOT the high-resolution 1D proton spectrum, but are instead the *projections* of the 2D data, and are therefore of much lower resolution, though probably still useful.

The automatic plotting routine does not know where to find the 1D spectrum, so it uses the projections instead. You can process the 2D data using the pages under the Process tab.

4.6 Interpreting the Calibration and Indanone Spectra

In this section, the data obtained from the automated probe calibration and 2% 2-ethyl-1-indanone sample are interpreted.

Calibration - When is it Necessary

Before acquiring spectra of a sample, the spectrometer must first be calibrated if one of the following is true:

- The probe has been changed
- A new probe has been installed
- The experiments are to be run at a different temperature than the last calibrations
- Solvent changes significantly (i.e., an organic lock solvent vs. deuterium oxide lock solvent)
- The calibration has not been run for several weeks (in this case it is simply a matter of good laboratory practice to check the calibration).

Interpretation of the Calibration Data

The first of the spectra, shown in Figure 3, is a ^{1}H observe with $CDCl_3$ as the lock solvent. This data is saved as `H1ref`. This spectrum contains three $^{13}CH_3I$ resonances centered at about 2.2 ppm. The center resonance is from the protons attached to carbon-12 and the two outer resonances are from the protons attached to carbon-13. The other resonances in the sample are from trimethylphosphite that has reacted with the methyl iodide.

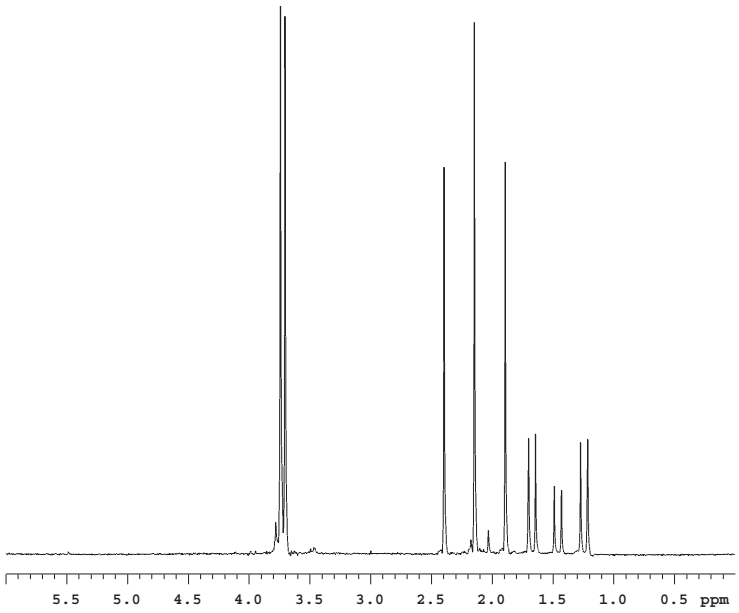


Figure 3. ^{1}H Spectrum of ^{13}C-Methyl Iodide

All trimethylphosphite has reacted to form a phosponate ester $(CH_3)P(=O)(OCH_3)_2$. This phosponate ester has a doublet at about 1.5 ppm, methyl group attached directly to ^{31}P and a triplet of doublets centered around 4 ppm that arise from ^{13}C (outer pair of doublets) and ^{12}C inner doublet of the methyl ester. The analysis of this sample is fully discussed in an article by Paul Keifer in *Magnetic Moments* (Keifer, P.A., *Magnetic Moments*, **1996**, 8 (#2), 18–20). The reaction results in a sample is partially enriched to give approximately 60% abundance of carbon-13 in methyl iodide. The natural abundance of carbon-13 is 1.1% so this level of enrichment is more then adequate for the purposes of calibration.

The next spectrum, shown in Figure 4, is an array of increasing ^1H pulse widths based on the ^1H pulse you specified in the Acquire window. If you did not enter a value for the pulse width it is set to the default targets `pw90` is set to 15 µs and `tpwr` of 51.

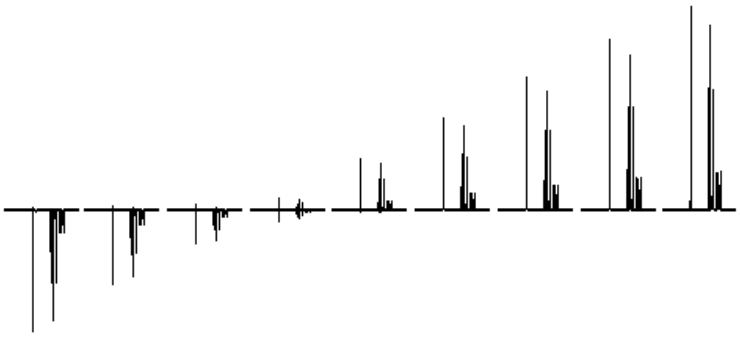


Figure 4. ^1H `pw` Array

The observe transmitter power is set to the value you specified and reduce by 3 for the first test. If the resulting `pw90` is shorter then the value you specified (or the default, if you did not specify a `pw90` target) the next test is started. If the `pw90` is longer then the target, the observe power is increased. Two attempts are made. If the calibration fails to achieve a `pw90` that is less than the specified `pw90` the AutoCalibration exits. If either attempt yields a `pw90` that is less then the specified value the AutoCalibration routine adjusts the observe power the remaining tests are aborted. If the `pw90` is less then the specified value, the AutoCalibration then adjusts the power until the measured `pw90` is no greater then the specified value but not more then 0.5 µs less than this value.The spectra from a successful calibration are saved as `H1pw90`.

The carbon `pwx90` calibration is the next experiment. The pulse sequence changes from `s2pul` to `PWXCAL`. The specification for the carbon `pw90` and `tpwr` are used as the target values. If no values were specified, the default values of 15 µs at power, in this case `pwxlvl`, of 51 are used as the target values. Just as with the proton `pw90` calibration, the AutoCalibration makes two attempts to achieve the specification and exits the AutoCalibration if the target specification is not reached after the second attempt. The data from the PWXCAL are saved as `C13pwx` and shown in Figure 5.

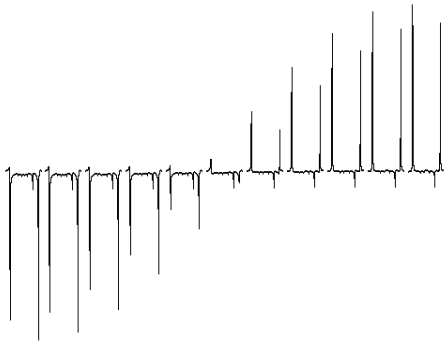


Figure 5. ^{13}C `pwx` Array

The next two experiments are run only if you have gradients. The first experiment calibrates the Z-gradient strength, produces the profile shown in Figure 6, and stores this information in the parameter `gcal`.

The next experiment calculates the ratios of the gradients to be used in various $^{1}H\{^{13}C\}$ indirect detection experiments and stores this information in the parameter `Cgrad` (for only ^{13}C), Figure 7.

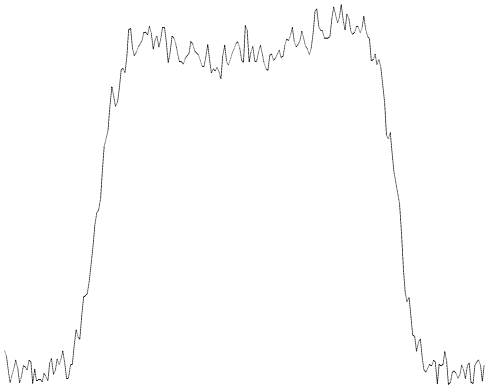


Figure 6. Gradient Profile

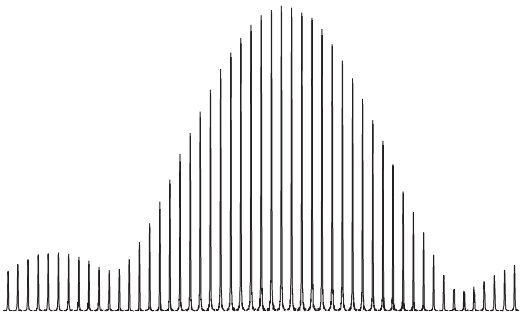


Figure 7. Gradient Calibrations

The next calibration is carbon observe pulse width and the pulse sequence is changed to `s2pul` for direct observation of the carbon. The calibration will follow the same pattern as the calibration of the proton `pw90` and the carbon `pwx90` using default values for target values if no target specification is given. A reference carbon spectrum is obtained first The full reference spectrum contains three sets of resonances, at the far right (approximately –22 ppm) is the ^{13}C resonance from methyl iodide, the doublet at 10 ppm is from the ^{13}C resonance from the phosponate methylester, and the 1:1:1 triplet (far left) at 78 ppm is the ^{13}C resonance of chloroform-d, $^{2}HCCl_3$. The carbon `pw90` calibration is analogous to the proton calibration. The reference carbon spectrum, shown in Figure 8, is saved as `C13ref`.

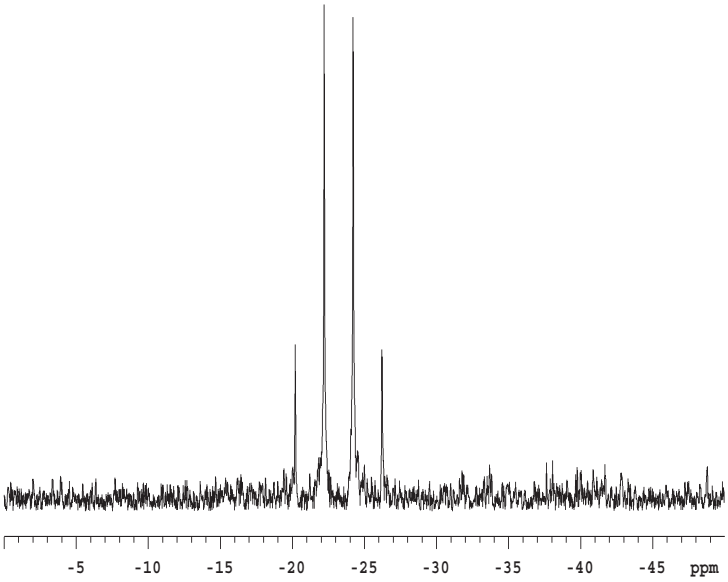


Figure 8. Proton Coupled ^{13}C Spectrum of ^{13}C-Methyl Iodide

The carbon observe pw90 is determined using a pw array, see Figure 9, and saved as C13pw90.

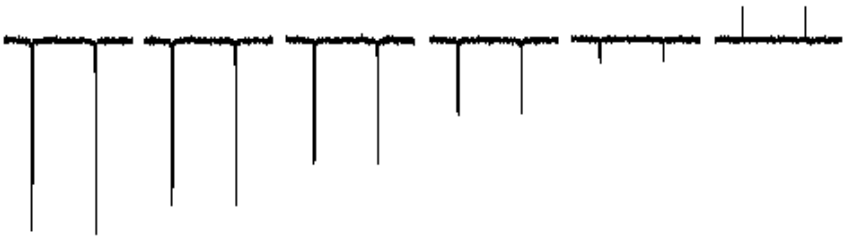


Figure 9. ^{13}C Observe pw Array of Proton Coupled Spectra

The final calibration is of the proton decoupler. The first calibration step determines the value of γH_2 at a decoupler power of 40 (the default value).

This measurement is made using continuous wave, cw, decoupling. The pulse sequence is the same as in the previous experiment, carbon observe with proton decoupling, except pw is now set to a fixed value, decoupler modulation mode; dmm is set to 'c', decoupler mode dm; is set to 'yyy', and the decoupler offset; dof is arrayed to produce the spectra shown in Figure 10.

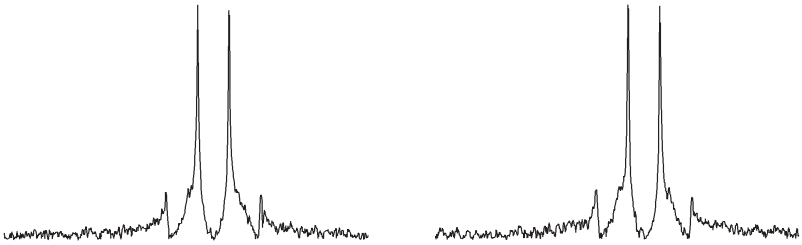


Figure 10. Proton Decoupler dof Array

From these spectra, the first estimate of the proton decouple pw90 is made. Using the relationship between the pulse width and the decoupler field strength, γH_2, shown in the equation below, the decoupler pw90 is determined.,

$$\gamma H2 = \frac{1}{4(pw90)}$$

The sequence is now set to ppcal and the proton decoupler 90° pulse, pp, is determined. These spectra, shown in Figure 11, are saved as Hdec_dept.

These parameters and calibrations are used to setup WALTZ decoupling. This completes the calibrations. During the calibration procedure, spectra and the array values are plotted to provide a permanent record of the calibrations.

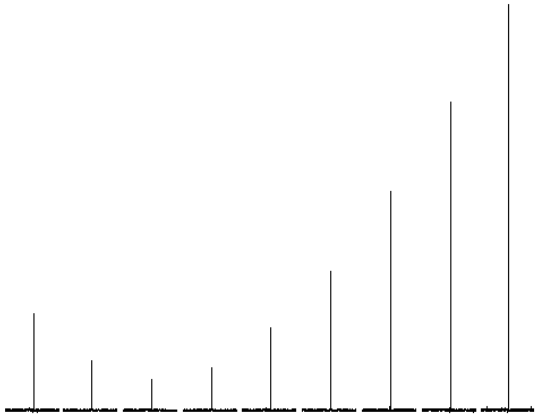


Figure 11. Calibration of the Decoupler 90° Pulse Width, pp

Interpretation of 2-Ethyl-1-Indanone Spectra

The proton NMR shows several distinct features. First, there are some impurities in the sample. These impurities, shown in Figure 12, are at the 2% level and some crosspeaks will show up in the 2D. The very large triplet for the methyl group has ^{13}C satellites at J=125 Hz. The singlet at 7.24 ppm is the residual $CHCl_3$ in the $CDCl_3$ solvent.

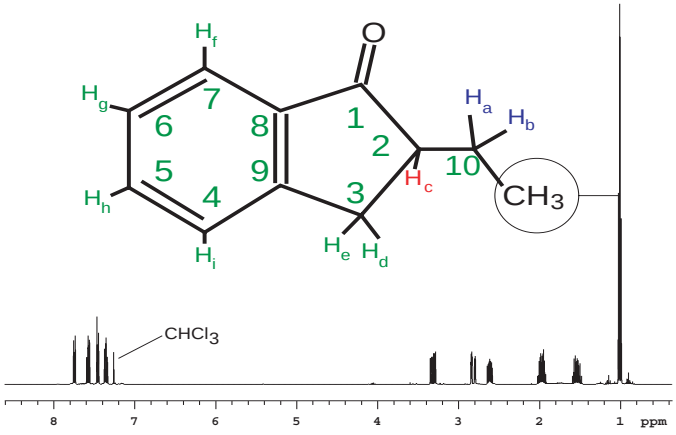


Figure 12. Proton Spectrum of 2-Ethyl-1-Indanone

The protons are assigned in the two expansions. The assignments are based upon the 2D data for the compound. The protons of the two CH_2 groups in the molecule, shown in Figure 13, are magnetically nonequivalent and show up as individual multiplets. The signals at 1.9 ppm and 1.55 ppm belong to protons on carbon 10. The two double doublets at 2.8 ppm and 3.26 ppm belong to protons on carbon 3. A complex multiplet at 2.6 ppm is the single proton on carbon 2

The protons of the aromatic ring, shown in Figure 14, are assigned based on the gHMBC and gCOSY data. Some minor impurities can be seen as well as the residual $CHCl_3$ signal. If the sample is shimmed very well there may be some truncation artifact on the $CHCl_3$ signal .

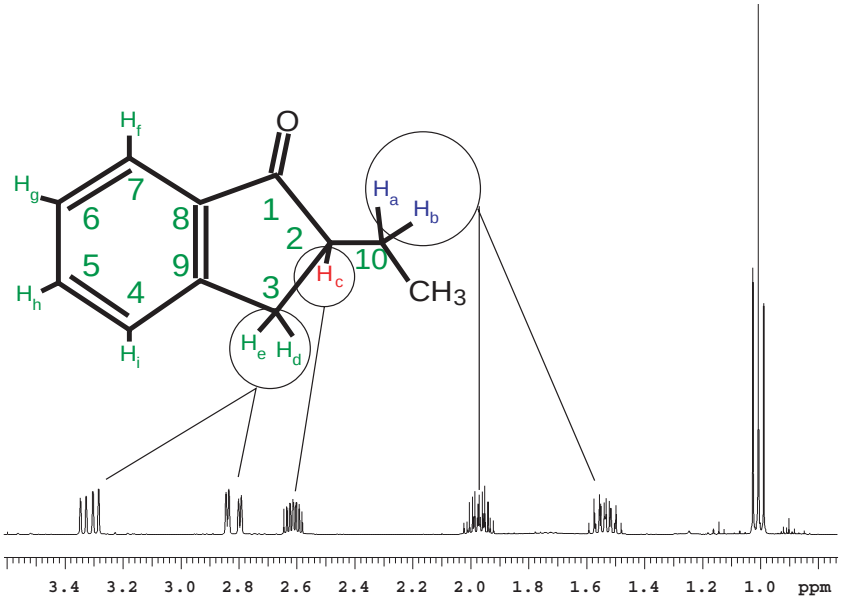


Figure 13. Aliphatic Region of the 2-Ethyl-1-Indanone Spectrum

.

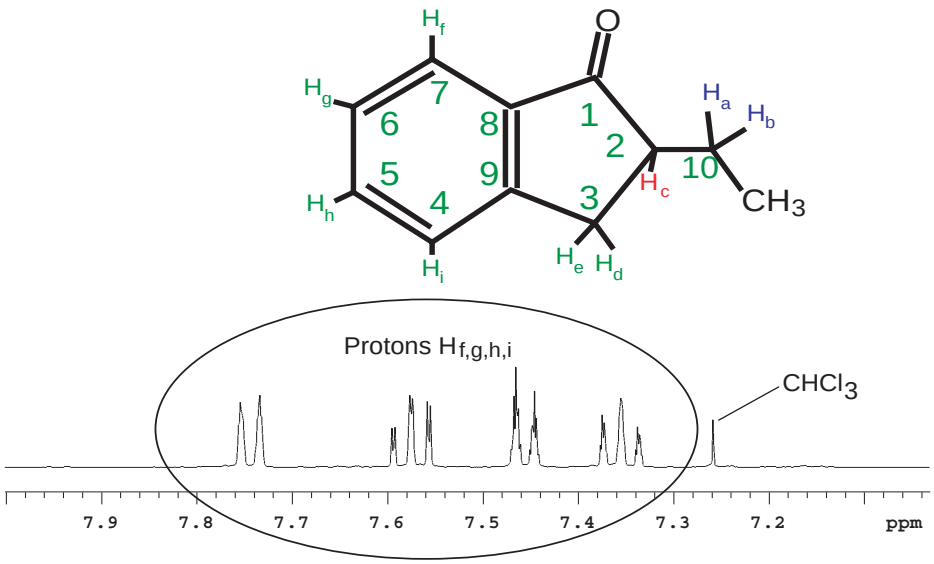


Figure 14. Aromatic Region of the 2-Ethyl-1-Indanone Spectrum

The gradient COSY shows cross peaks describing the coupling pathways. Some smaller cross peaks are also present in the spectrum which actually arise from the impurities shown in Figure 15. An example of this is the cross peak at 3.5 ppm

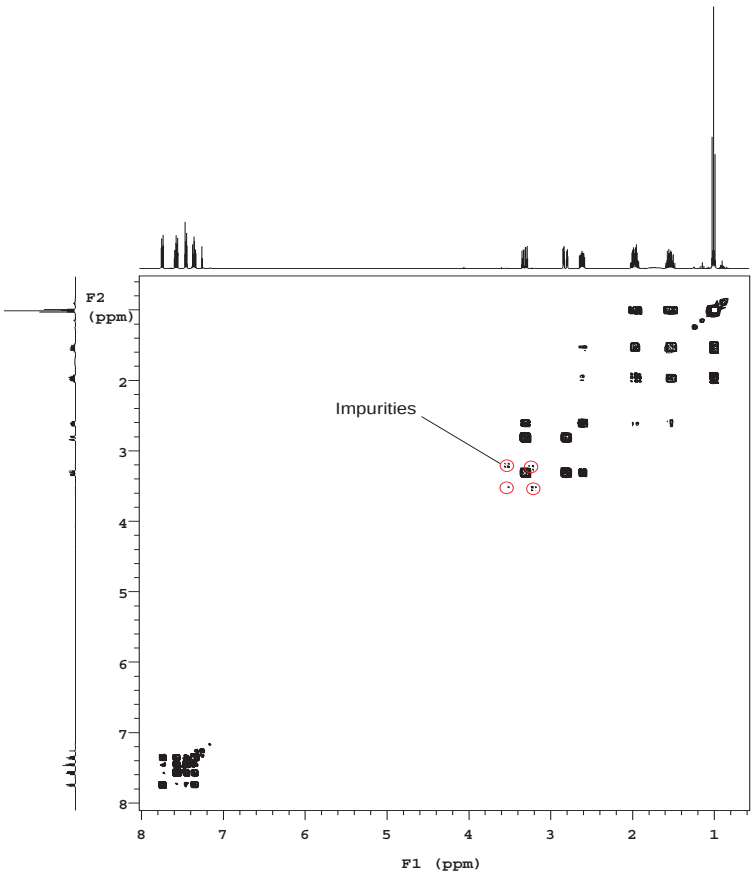


Figure 15. Gradient COSY of 2-Ethyl-1-Indanone

The methyl triplet in Figure 16 shows major cross peaks to the H10 protons. The multiplicity of the H10 and H2 protons cross peaks show that they are weakly coupled (the J value is small).

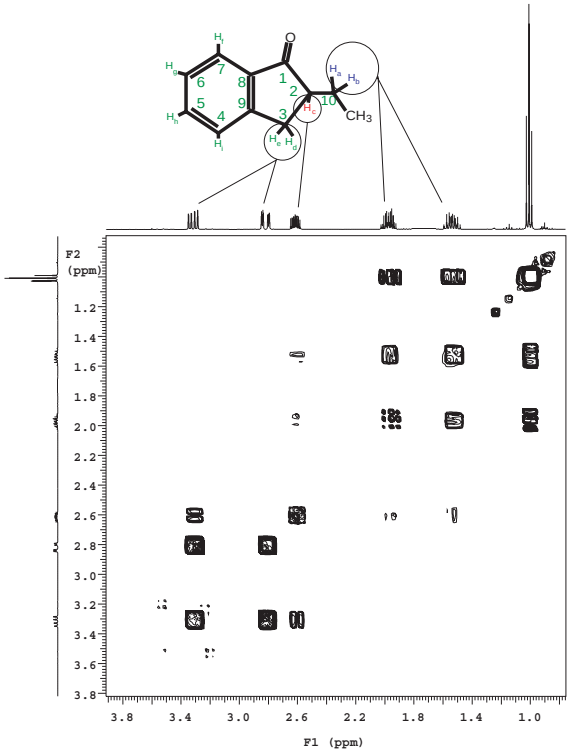


Figure 16. Gradient COSY (gCOSY) of Aliphatic Region of 2-Ethyl-1-Indanone

Assignment of the aliphatic region, begins with H7, the most deshielded proton, Figure 17. From H7 direct connectivity is apparent to H6. The rest of the assignment is H6 to H5 (the other triplet) and then to H4.

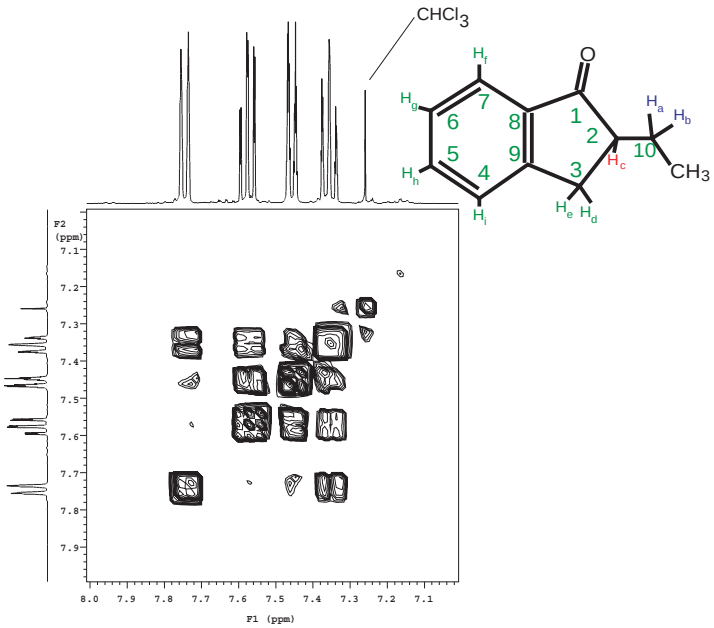


Figure 17. Gradient COSY (gCOSY) of the Aromatic Region of 2-Ethyl-1-Indanone

The assignment of H7 to the signal at 7.72 ppm is confirmed by the gHMBC data.

TOCSY is a phase sensitive experiment. The cross peaks are narrower than in the COSY giving higher "resolution". Correlations among all protons in a spin system are observed in the TOCSY spectrum, see Figure 18. The critical parameter is mix. In this case mix is 0.08 seconds which is sufficient to show correlations throughout the entire spin system. Shorter mix times will reveal fewer correlations.

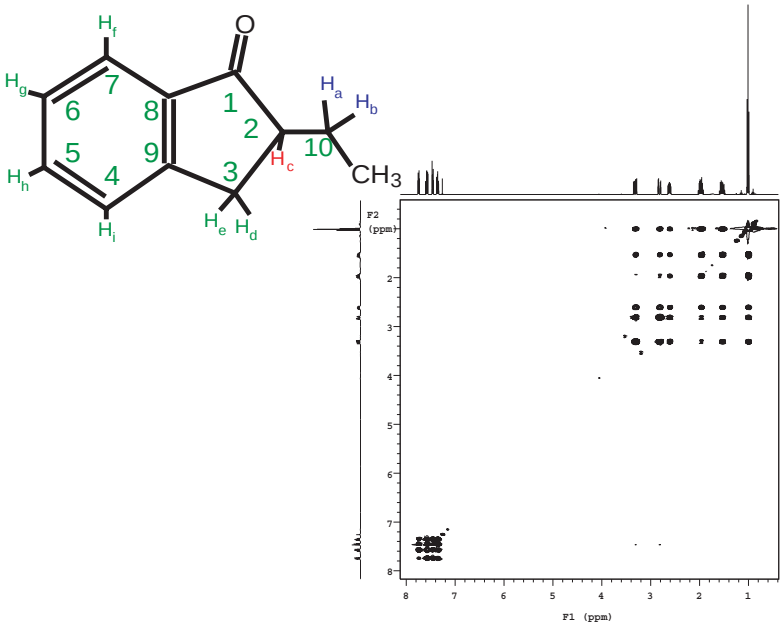


Figure 18. TOCSY of 2-Ethyl-1-Indanone shows Correlations Among All Protons

The expansion shows the completely defined spin system starting with the CH_3 group and ending with protons on C10, Figure 19. A total of 5 crosspeaks are seen in the row]

The indanone sample does not have any significant NOE crosspeaks, Figure 20. The main area of interest in this spectrum is to note that the diagonal will be negative and the NOESY crosspeaks will be positive. Crosspeaks which appear to have both positive and negative components are actually not NOE correlations but coupling artifacts.

In the gHSQC (and HSQC) experiment, see Figure 21 the protons correlate with the carbons to which they are attached. The detected nucleus is ^{1}H and this results in a higher signal-to-noise then the ^{13}C detected hetcor experiment. When compared to the HMQC experiment, the HSQC experiment has the advantage that the $^{1}H - {}^{1}H$ homonuclear coupling do not evolve. As a result the resolution in the 2D plane is higher in the HSQC experiment. The higher resolution has the added advantage of improving the signal-to-noise. The version of the HSQC experiment supplied with VnmrJ has the added benefit that it will distinguish –CH, $–CH_2$, and $–CH_3$ groups. In this case phase is indicated by whether the crosspeak is filled in with multiple contours (above the plane) or is a single contour (below the plane).

By contrast to the gHSQC experiment, the gHMBC (and HMBC) experiment shows long range (mostly 2 and 3 bond) $^{1}H - {}^{13}C$ correlations. This shows connectivity between the non - protonated carbonyl and the protons on C-2.

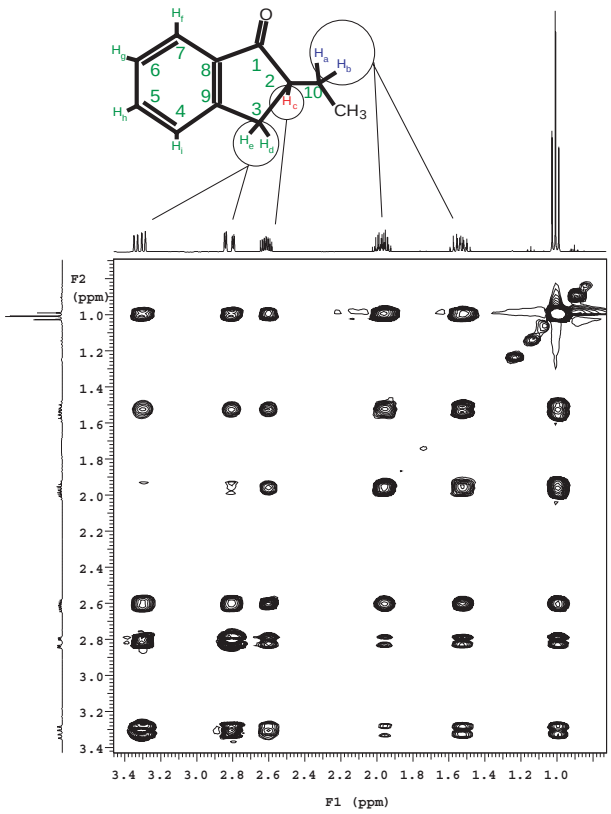


Figure 19. TOCSY of 2-Ethyl-1-Indanone, Correlations of Protons on C11, C10, C3

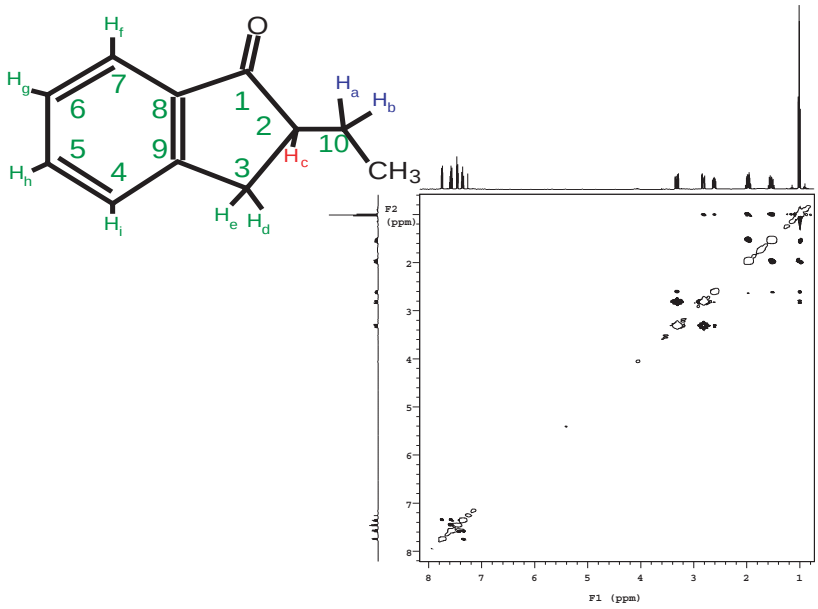


Figure 20. NOESY Spectrum of 2-Ethyl-1-Indanone

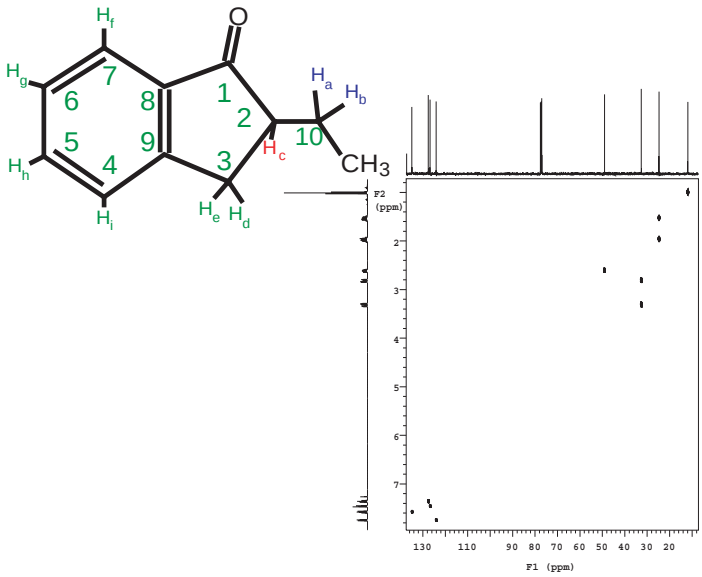


Figure 21. HSQC Spectrum of 2-Ethyl-1-Indanone

4.7 Magnet Maintenance

Magnet maintenance is described in the *VnmrJ Installation and Administration* manual. Before undertaking any work with the magnet, read the warnings below and in the "SAFETY PRECAUTIONS," page 5 of this manual. Also, be sure to review the documentation provided by the magnet vendor.

WARNING: **The extremely low temperature of liquefied helium and nitrogen can cause skin damage similar to high-temperature burns. Contact with the cold gas evolving from the liquid may produce the same effect. Delicate body tissues, such as the eyes, are easily damaged by exposure to cold gas or liquid. Skin can stick to metal that is refrigerated by liquid helium and can tear when pulled away. Immediately flood with large quantities of unheated water any area of the body that is "burned" by liquid or cold gas, and then apply cold compresses. If the skin is blistered or there is any chance the eyes are affected, immediately seek medical treatment.**

WARNING: **Wear goggles and loose-fitting protective gloves while working with cyrogens.**

Magnet maintenance consists of three basic elements:

- Periodic checks of the cryogen levels and boil off rates
- Liquid nitrogen fill
- Liquid helium fill.

All cryogens should be delivered in *nonmagnetic* dewars. Consult the manual supplied by Oxford complete instructions and for more detailed information on cyrogen fill intervals and capacities.

CAUTION: **Failure to maintain the correct liquid nitrogen levels will lead to excessive liquid helium boil off which in turn may lead to a quench of the magnet.**

CAUTION: **Failure to maintain the correct liquid helium levels may lead to a quench of the magnet.**

4.8 30-Day System Maintenance

The following procedures must be performed while logged in as `vnmr1`.

1. Updating Solvent Shims

   Reshim lineshape and then save shims to `/vnmr/shims/acetone`.

2. Check Calibration of `pw90` and `tpwr`, as described in "Calibrating a Probe," page 15.

3. Perform hard drive maintenance: delete unused files, directories, old FIDs, macros, shims, or users.

4. Check magnet drift, as described in 2.3 "Magnet Drift Test" page 23. If necessary (when Z0 is high or close to maximum 2047, reset Z0 by adjusting the `lockfreq` parameter as follows:

   a. Load **`H1sn.par`**.

b. Set **pw=1**, **gain=2**, **d1=0**, **in='n'**, **at=1**, **sw=100**.

c. Set Z0 to 0.

d. Insert a pure water sample and acquire a spectrum.

e. Enter **f full aph0**.

f. Place a cursor on the water peak and enter **nl movetof setlockfreq**.

4.9 Warranty and Who to Call for Assistance

The installation engineer will explain the standard warranty terms. Non standard warranty terms, if included as a condition of sale, are detailed in the sales contract and will not be covered by the installation engineer.

Online Customer Support and Information	**Webb Addresses**	
Service and Technical Support	http://www.varianinc.com/nmr/service	
Applications Support	http://www.varianinc.com/nmr/apps	
Sales Support	http://www.varianinc.com/nmr/contact	
Product Information	http://www.varianinc.com/nmr/products	
In warranty Service	**Location**	**Phone Number**
United States and North America	Palo Alto, California	1.800.356.4437
Europe	Local Sales Office	See "Varian Sales Offices"
Other International	Local Sales Office	See "Varian Sales Offices"
Technical Support	**Location**	**Phone Number**
United States and North America	Palo Alto, California	1.800.356.4437
Europe	Oxford, UK	44.1865.388.800
Other International	Local Sales Office	See "Varian Sales Offices"
Post Warranty Support	**Location**	**Phone Number**
United States and North America	Palo Alto, California	1.800.356.4437
Europe	Local Sales Office	See "Varian Sales Offices"
Other International	Local Sales Office	See "Varian Sales Offices"
Applications Support	**Location**	**Phone Number**
United States and North America	Palo Alto, California	650.424.4526
	Columbia, Maryland	410.381.7229
Europe	Darmstadt, Germany	49.6152.703.253
Japan	Tokyo, Japan	81.3.5232.1211
Other International	Local Sales Office	See "Varian Sales Offices"

Varian Sales Offices

For product sales and service information, contact one of the Varian sales offices:

- Argentina, Buenos Aires, (114) 783-5306
- Australia, Mulgrave, Victoria, (3) 9566-1138
- Austria, Vösendorf, (1) 699 96 69
- Belgium, Brussels, (02) 721 51 51
- Brazil, Sao Paulo, (11) 829-5444

- Canada, Ottawa, Ontario, (613) 260-0331
- China, Beijing, (10) 6846-3640
- Denmark, Herlev, (42) 84 6166
- France, Orsay, (1) 69 86 38 38
- Germany, Darmstadt, (6151) 70 30
- Italy, Milan, (+39) 02 9273401
- Japan, Tokyo, (3) 5232 1211
- Korea, Seoul, (2) 3452-2452
- Mexico, Mexico City, (5) 523-9465
- Netherlands, Houten, (0118) 61 71 56
- Norway, Oslo, (9) 86 74 70
- Russian Federation, Moscow, (95) 241-7014
- Spain, Madrid, (91) 472-7612
- Sweden, Solna, (8) 445 1601
- Switzerland, Zug, (41) 749 88 44
- Taiwan, Taipei, (2) 2698-9555
- United Kingdom, Walton-on-Thames, England (1932) 898 000
- United Kingdom, Oxford, England
  Sales and Service +44(1865) 388 883
  E- mail technical.support@varianinc.com
  NMR Service Manager, Europe and Pacific Rim
- United States, Palo Alto, California,
  Varian, Inc., NMR Systems
  Customer Sales Support, (650) 424-5434
  Service Support, Palo Alto, California, **1 (800) 356-4437**
  E- mail: nmr.customersupport@varianinc.com
  North American Service Manager
  9017 Mendenhall Ct., Ste D, Columbia, MD 21045
  (410) 381-7229
- Venezuela, Valencia (41) 257608

Chapter 5. Acceptance Test Results

This chapter contains the following forms for recording system information and acceptance test results:

Notes:

5.1 Computer Audit

Information about your site (please print):

Company/University _____

Address _____

_____

Principal User _____

Phone _____ Spectrometer type _____

Fax _____ Console S/N _____

Sales Order No. _____ Delivery (month/day) _____

Information on each computer (additional forms are on the back of this page). Include computers directly attached to the spectrometer, computers (networked or non-networked, on-site or off-site) used to process NMR data using Varian's VnmrJ software, and computers (on-site and off-site) used to process data collected on this spectrometer with software from other vendors.

Information on computer ____ of ____ (e.g., 1 of 3)

Manufacturer _____ Model no. _____

Computer S/N _____ Purchased from _____

Memory (Mbytes) _____ Screen size (in.) _____

Peripherals: Internal hard disk (Mbytes) _____

External hard disk (Mbytes) _____ Serial no. _____

Tape drive size _____ Serial no. _____

CD-ROM drive model _____ Serial no. _____

Printer model _____ Serial no. _____

Plotter model _____ Serial no. _____

Terminal model _____ Serial no. _____

Other peripheral _____ Serial no. _____

Computer function: NMR host _____

Workstation running VnmrJ _____ on-site or off-site

Workstation running other NMR software _____ on-site or off-site

Workstation running VnmrJ and other NMR software _____ on-site or off-site

VnmrJ version _____ **Operating system** _____

The above computer audit was performed during installation of the system.

_____

Varian Representative Date

I certify that the information on this form is accurate and that all computers to be used to run VnmrJ software (including variants VnmrS, VnmrX, VnmrI, VnmrSGI, and VnmrV), or to run other software to process data obtained on this spectrometer, have been included in the audit (including those previously registered as part of purchases of other Varian NMR spectrometers).

_____

Customer Representative Date

Use these forms for additional computers. If more forms are needed, copy this page. Attach all copies to the Computer Audit.

Information on computer ____ of ____ (e.g., 2 of 3)

Manufacturer _____ Model no. _____

Computer S/N _____ Purchased from _____

Memory (Mbytes) _____ Screen size (in.) _____

Peripherals: Internal hard disk (Mbytes) _____

External hard disk (Mbytes) _____ Serial no. _____

Tape drive size _____ Serial no. _____

CD-ROM drive model _____ Serial no. _____

Printer model _____ Serial no. _____

Plotter model _____ Serial no. _____

Terminal model _____ Serial no. _____

Other peripheral _____ Serial no. _____

Computer function: NMR host _____

Workstation running VnmrJ _____ on-site or off-site

Workstation running other NMR software _____ on-site or off-site

Workstation running VnmrJ and other NMR software _____ on-site or off-site

VnmrJ version _____ **Operating system** _____

Information on computer ____ of ____ (e.g., 3 of 3)

Manufacturer _____ Model no. _____

Computer S/N _____ Purchased from _____

Memory (Mbytes) _____ Screen size (in.) _____

Peripherals: Internal hard disk (Mbytes) _____

External hard disk (Mbytes) _____ Serial no. _____

Tape drive size _____ Serial no. _____

CD-ROM drive model _____ Serial no. _____

Printer model _____ Serial no. _____

Plotter model _____ Serial no. _____

Terminal model _____ Serial no. _____

Other peripheral _____ Serial no. _____

Computer function: NMR host _____

Workstation running VnmrJ _____ on-site or off-site

Workstation running other NMR software _____ on-site or off-site

Workstation running VnmrJ and other NMR software _____ on-site or off-site

VnmrJ version _____ **Operating system** _____

5.2 Installation Customer Training Checklist

Customer Information

Company/ University _____

Address _____

_____

Principal User _____

Phone _____ Spectrometer type _____

Fax _____ Console S/N _____

Sales Order No. _____ Magnet S/N _____

Magnet Familiarization:

Done	*Topic*	*Reference*
	Overall magnet familiarization	Oxford Magnet Reference Manual
	LHe and LN_2 top off procedures	Oxford Magnet Reference Manual and Lhe refill video
	Use of flow meters	Oxford Magnet Reference Manual
	Antivibration system operation	Antivibration Accessory Installation Manual
	Posting of magnetic field warning signs	Appendix A, "Posting Requirements for Magnetic Field Warning Signs,"

Probes:

Done	*Topic*	*Reference*
	Probe installation removal, tuning, and filter setup for ID and TR experiments	Choose a specific probe manual AutoSwitchable probes Broadband probes Indirect Detection probes Nano probes Dual and DualBB probes Flow probes
	VT system installation	Variable Temperature Unit Installation
	VT system operation	User Guide: Liquids NMR

Console:

Done	*Topic*	*Reference*
	Major component overview	System Overview
	OS Shut down procedures	Solaris Installation and Administration
	Host console connection and overview	VnmrJ Installation and Administration

Host Computer:

Done	*Topic*	*Reference*
	Host computer setup	Solaris or Linux
	VnmrJ installation	VnmrJ Installation and Administration
	Directory structure overview	VnmrJ Installation and Administration
	Using acqproc and makeuser commands	VnmrJ Installation and Administration
	Storage devices and accessories	Solaris Administration

Spectrometer Operation:

Done	*Topic*	*Reference*
	Standard parameter and probe calibration	Walkup NMR
	Creating a composite protocol	Walkup NMR
	Interpretation of Indanone spectra	Walkup NMR
	Manual lock and shim demonstration	VnmrJ Liquids NMR
	Basic spectral display procedures	VnmrJ Liquids NMR
	Gradient shimming demonstration	VnmrJ Liquids NMR

Miscellaneous:

ATP signoff and Walkup NMR demo, if applicable

Hallmark of Quality

Customer contacts for service, applications etc.

Accessory training as required

Varian Representative Date

Customer Representative Date

5.3 System Installation Checklist

Company/University _____

Address _____

_____

Principal User _____

Phone _____ Spectrometer type _____

Fax _____ Console S/N _____

Sales Order No. _____ Magnet S/N _____

Shipment Damage:

_____

_____

Preinstallation Preparation:

Line voltage measured (Vac): console _____ accessory _____

Line pressure: air _____ N_2 _____

Air conditioning: _____

Cryogens (liters): LHe _____ LN _____

Testing:

1. Acceptance tests and computer audit
   - ___ Acceptance tests procedures finished
   - ___ Test results form completed and signed
   - ___ Computer audit completed and signed
2. System documentation review

   Software Object Code License Agreement (acceptance of product constitutes acceptance of
   - ___ object code license regardless of whether agreement is signed or not)
   - ___ Varian and OEM manuals
   - ___ Explanation of warranty and where to telephone for information
3. Installation Training
4. Installation Customer Training Check list

Notes:

5.4 Supercon Shim Values

Fill in the following information:

Magnet Frequency and Serial Number

Magnet Frequency _____

Serial Number _____

Measurement in

Helipot _____

Amps _____

Measurements

Measurement	1. Date:	2. Date:	3. Date:
Z0			
Z1			
Z2			
Z3			
Z4			
X			
Y			
ZX			
ZY			
XY			
X2–Y2			
Drift			
Boiloff (N2, He)			
Spacers			
Main Field Current			
Customer Signature:			
Varian Representative Signature:			

Notes:

5.5 Console and Magnet Test Results

Fill in the following information:

From 2.1 “Calibrating a Probe” page 15

From 2.2 “Automated Data Acquisition” page 19

From 2.3 “Magnet Drift Test” page 23

From “Variable Temperature Operation (Optional Hardware)” page 25

From “Temperature Accuracy for VT Systems (Optional Test),” page 26

From “Stability Calibration for High-Stability VT (Optional Test),” page 29

Varian Representative Date

Customer Representative Date

Notes:

Appendix A. Posting Requirements for Magnetic Field Warning Signs

The strong magnetic fields that surround a superconducting magnet are capable of causing death or serious injury to individuals with implanted or attached medical devices such as pacemakers or prosthetic parts. Such fields can also suddenly pull nearby magnetic tools, equipment, and dewars into the magnet body with considerable force, which could cause personal injury or serious damage. Moreover, strong magnetic fields can erase magnetic media such as tapes and floppy disks, disable the information stored on the magnetic strip of automated teller machine (ATM) and credit cards, and damage some watches.

To warn of the presence and hazard of strong magnetic fields, the customer is responsible for posting clearly visible signs warning of magnetic field hazards. This responsibility includes measuring stray fields with a gaussmeter.

Radio-frequency emissions may also pose a danger to some individuals. The rf emission levels from Varian NMR equipment have been measured and compared to the IEEE/ANSI C95.1-1991 standard. For further information, refer to the *RF Environment* section of the *Installation Planning Guide*.

Warning Signs

Varian provides signs to help customers meet this posting responsibility. These signs *must* be posted according to the following requirements *before the magnet is energized*:

1. *10-gauss warning signs* (Figure 22) – Post along the 10-gauss perimeter of the magnet so that a sign can be easily seen by any person about to enter the 10-gauss field from any direction. Refer to the manuals supplied with the magnet for the size of a typical 10-gauss stray field. Check this gauss level after the magnet is installed.

   Note that the stray field may extend vertically to adjacent floors, and additional signs may be needed there. A sign is not required if the 10-gauss field extends less than 30 cm (12 in.) beyond a permanent wall or less than 61 cm (24 in.) beyond the floor above the magnet.

2. *5-gauss warning signs* (Figure 23) – Post along the 5-gauss perimeter of the magnet so that a sign can be easily seen by any person about to enter the 5-gauss field from any direction. Refer to the manuals supplied with the magnet for the size of a typical 5-gauss stray field. Check this gauss level after the magnet is installed. Note that the stray field may extend vertically to adjacent floors, and additional signs may be needed there.

3. *Magnet area danger signs* (Figure 24) – Post at each entrance to the magnet area. Be sure each sign is outside the 5-gauss perimeter.

Figure 22. 10-Gauss Warning Sign

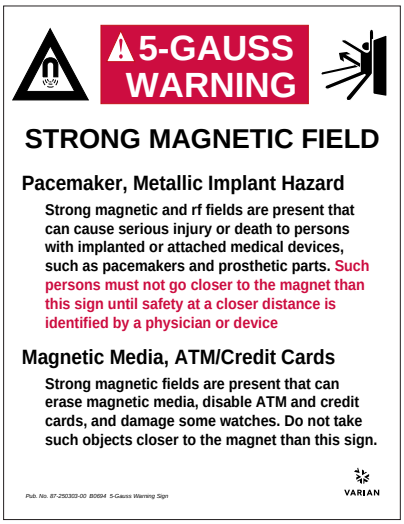


Figure 23. 5-Gauss Warning Sign

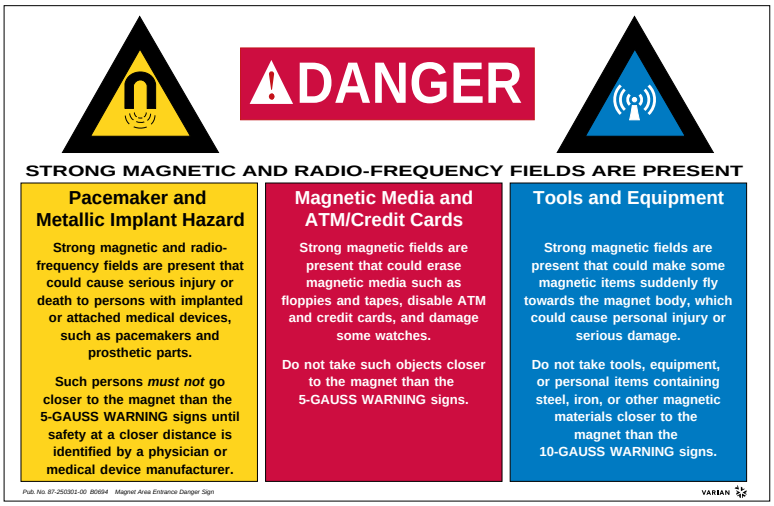


Figure 24. Magnet Area Danger Sign

Stray magnetic fields can reach beyond the published distances when two or more magnetic fields intersect or when the field extends over large ferromagnetic masses or structures (steel doors, steel construction beams, etc.). In this case, the customer *must* measure the stray field using a gaussmeter to determine how the 5- and 10-gauss fields are altered (contact a scientific instrumentation supplier for information on acquiring a gaussmeter).

You can request additional signs from Varian by telephoning 1-800-356-4437 in the United States or by contacting your local Varian office in other countries.

Numerics

A

B

C

D

E

F

G

H

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L

M

N

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