

Walkup NMR Quick Guide

Use this page to quickly reference the order of operations on the walkup NMR instruments. For more information on any of the steps, see the detailed walkthrough or contact the MRC staff.

1. Schedule time on the MRC scheduling calendar.
2. Login to the instrument using your NMR username and password.
3. Eject the standard and insert your sample.
4. Enter a sample name (optional) and select the solvent (required).
5. Lock the sample using **Auto Lock** or by manually locking.
6. Shim the sample (maximize the lock level using the shims). Shim order: Z1, Z2, Z1
7. Verify the lock power is less than the lock gain and set the lock level between 30 and 70.
8. Select the desired experiment from the Experiment Selector.
9. Adjust parameters under the Acquisition tab. Make sure Auto Gain is checked.
10. Start the experiment with `ga` or click the **Go** button.
11. Process the data. You will need to at least transform the data (`wft`) before saving/emailing.
 - i. Most processing can be completed using the **Auto process** button on the **Process** tab.
12. (Optional) Plot your spectrum.
13. Save and/or email your data.
14. Continue with your next experiment. (Step 7)
OR
Change to your next sample. (Step 2)
15. If finished, check the height of the standard D₂O sample and insert it into the magnet.
16. Logout and complete the logbook with any missing information.

Walkup NMR Walkthrough

Notes on sample prep:

All sample preparation is to take place in your wet lab. No preparation equipment is permitted in the NMR labs without prior permission from the MRC staff. If you need to bring anything other than NMR tubes to the lab, consult the MRC staff before scheduling time. Gloves and lab coats are not permitted in the NMR labs. Remove any gloves at the door before using the instruments.

Samples should be free of particulates (filtered) and non-viscous for best lineshapes. Make sure your sample height is 5 cm tall (about 0.6-0.7 mL solvent) and is sealed **only** with a plastic cap.

Do not use parafilm, foil, or PTFE tape (alone or wrapped around a plastic cap) to cap your samples.

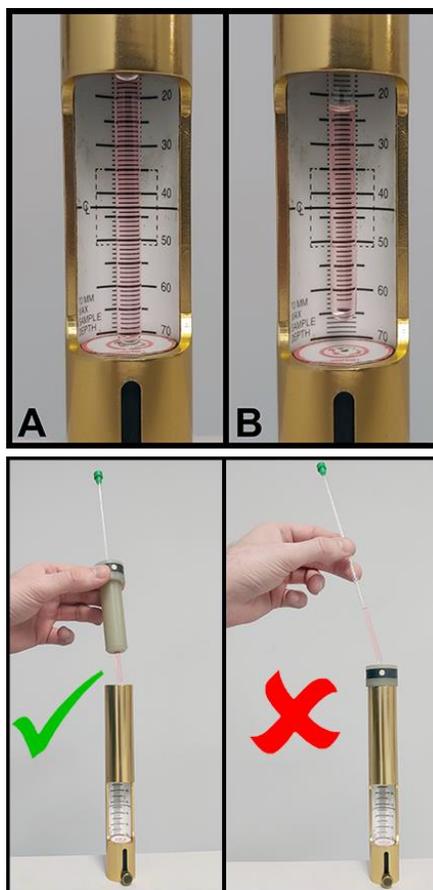
1. Place any ferromagnetic items on the desk (watch, keys, bracelets, etc). This includes metal objects such as buckles if freely hanging from your clothing.

Sign into VnmrJ near the bottom of the screen with your NMR username and password.

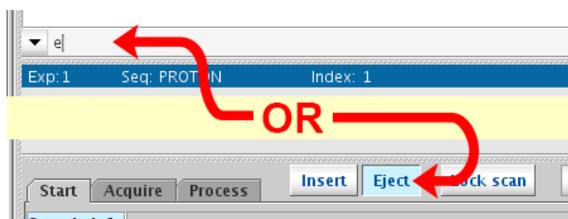


2. Load your sample into a spinner and use the depth gauge to set the sample height. You will need to raise the tube in the spinner for shorter samples (B). Samples 5 cm in height or greater should be inserted fully into the sample depth gauge (A).

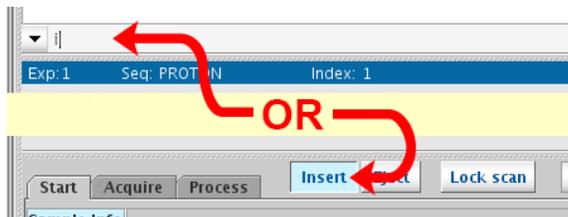
Do not place the spinner into the depth gauge before loading your sample. This risks breaking your NMR tube. Load samples into spinners by hand first; making sure your hand is not more than 10 cm from the top of the spinner to avoid snapping your tube. Avoid touching the sample area of the tube to prevent finger oils from showing in the spectra.



3. Select the **Start** tab if not already selected. Click the **Eject** button or enter `e` on the command line. Take out the sample in the magnet and place it in the sample rack.

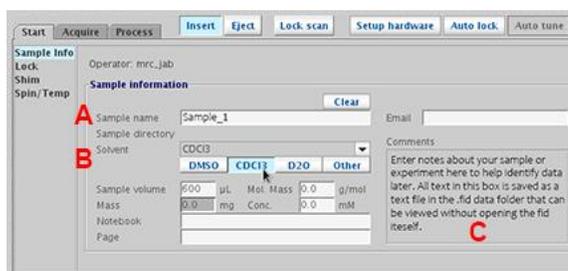


4. Insert your sample into the magnet bore. Make sure the sample will float at top before releasing it. Click the **Insert** button or enter `i` on the command line.



5. Enter a sample name (**A**, optional) and select the solvent (**B**, required) on the **Sample Info** page. Use the drop-down menu to select a solvent other than DMSO, CDCl₃, or D₂O.

It is recommended to enter basic information about the sample in the Comments box (**C**), which will help you keep data organized.



6. Click the **Auto Lock** button. After the process runs, wait for the lock level to stabilize and regulate. If Auto Lock fails, a message will appear in the top command window and you will need to lock manually (Step 7). Otherwise skip to Step 8.

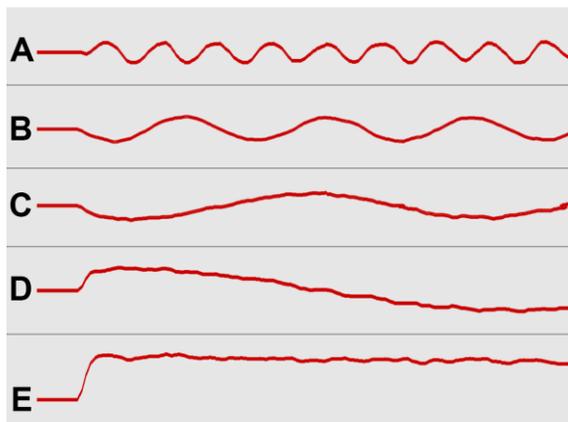


7. Manual locking

- Select the **Lock** page
- Uncheck the **Lock** checkbox to turn off lock.

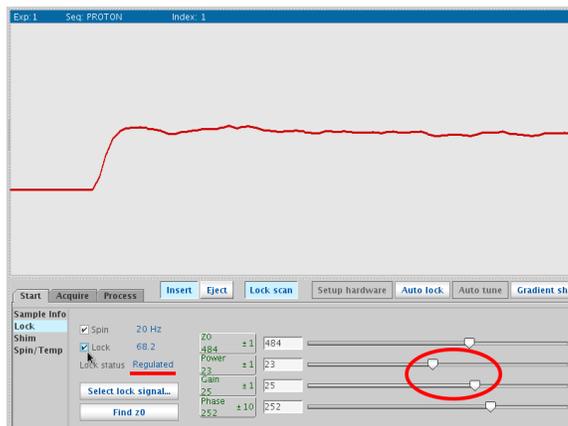
! Lock must be off before adjusting **Z0**.

- Click **Lock scan**.
- Adjust **Z0** to obtain zero beat frequency (**E**, on-resonance, 'stairstep' shape line). If starting far from the on-resonance value, you occasionally will see a sine wave with multiple periods (**A**). To get lock, move **Z0** such that you decrease the number of periods seen (**B**→**D**) until you reach **E**. The value can be increased by right-clicking on the **Z0** button or decreased by left-clicking. The middle click will adjust the step size between 1, 10, and



100. Specific values may be typed into the text box next to the button. You must press Enter after typing the number to activate it.

- If necessary, adjust **Gain** to keep the lock level in the middle of the screen while adjusting **Z0**. The **Gain** slider should always remain above the **Power** slider. Do not continue adjusting **Z0** if the line reaches the top of the screen (Lock Level > 80). Reduce the **Power** and/or **Gain** first, and then continue. If **Gain** reaches the maximum, you can increase **Power**, but it should not be increased past 40 except for low sample sizes or low deuterium-concentrated solvents. Do not adjust the **Phase** parameter yet.
- Click the **Lock** checkbox to turn on and regulate lock.



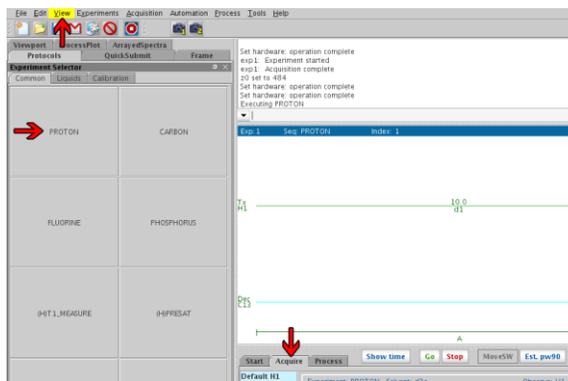
8. Shimming

- Select the **Shim** page and adjust the **Lk Power** and **Lk Gain** if necessary so the lock level is around 30-50.
- On the 300: Adjust **Z1C** to maximize the lock level and then adjust **Z2C** (both highlighted in yellow).
- On the 300 and 400: Adjust **Z1** and **Z2** (highlighted in green) in the same manner as above to make fine adjustments to the lock level.
- If at any time the Lock level maxes out (≥ 100), reduce Lk Power and Lk Gain together to lower the Lock value < 80 and continue.
- Adjust **Lk Phase** to maximize the lock level.
- Adjust **Lk Power** and **Lk Gain** to set the Lock Level between 30 and 70. Verify that the **Gain** slider on the Lock page is above the **Power** slider and adjust as needed.



9. Running an Experiment

- Select the **Acquire** tab
- Select an experiment from the left-hand side (e.g. PROTON, CARBON, etc). If the buttons are missing, look under the **Protocols** tab or select **Experiment Selector** from the **View** menu.



- ## 10. The Default [Nuclei] page contains the most frequently adjusted parameters and common values for each. All these values should be verified before running. If a desired value is not present in the drop down, custom values may be entered in the corresponding field on the **Acquisition** page.

Verify that the **Autogain** checkbox is selected on the **Default** page.



General Notes:

- The default instrument parameters are set to obtain survey spectra with the optimum signal-to-noise in the shortest time for average MW. They usually work for proton counting (i.e. distinction between CH, CH₂ and CH₃ proton resonances), but are **NOT suitable for quantitative analysis**. The user can modify the parameters to get desired results according to their project needs.
- For accurate quantitative analysis, parameters must be altered (e.g. for quantitative ¹H spectra pw(90) and d1=5xT₁; for quantitative ¹³C spectra pw(90), d1=5xT₁ and dm='nny'). You can guess T₁ based on prior knowledge from your group's work or published results with similar samples. However, you should perform a T₁ measurement experiment and use identical sample preparation conditions (solvent, analyte, concentration, temperature) if you wish to use that measured T₁ as a basis for quantitative analysis of future samples. You should consult the MRC staff to ensure proper experimental setup before running quantitative experiments if you plan to publish the results.
- If you work with atypical structures (e.g. paramagnetic materials, anions or cations) the default spectral window might not be suitable.
- Data processing can dramatically influence the reliability of quantitative analysis. For best results, contact the lab staff for instructions on quantitative data processing.

- Signal-to-noise can be improved with larger number of scans (Scans requested, nt). Always use nt equal to a multiple of 4 to avoid artifacts in the spectra.
- The **Show time** button will display the length of the experiment in the command window.
- Line broadening (lb) and transform size (fn) can be set before acquiring data, but can also be set while processing (which is why you'll find them under the **Processing** tab). It is recommended to set the transform size to 2X the number of points to improve lineshape. You can quickly do this by entering $fn=2*np$ on the command line. Recommended values of lb are 0.5 or 1 for 1H and 1 or 2 for ^{13}C to improve the signal-to-noise.

11. Start the experiment by clicking the **Go** button or entering ga on the command line. Use the **Stop** button or enter aa on the command line if you need to end the experiment for any reason.



Leaving the instrument unattended

You may leave an experiment running without placing an 'experiment in progress' sign at the instrument. Fill out the logbook and include your phone number and solvent(s) before leaving. The screen will lock after 10 minutes or you can click the **Lockscreen** button in the **Start** tab.

Data Processing

Your data should automatically process at the end of the experiment. Steps 12-17 will walk through basic data processing in VnmrJ. You may skip to **Step 19** if you prefer to just save your data and process it later.

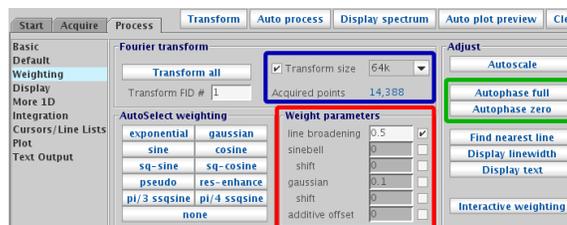
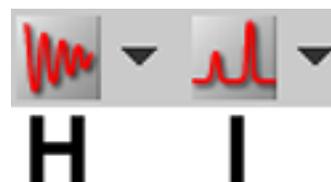
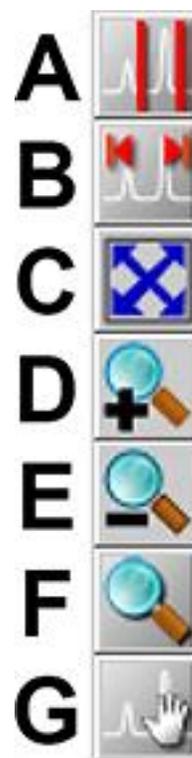
12. Select the **Process** tab and click the **Auto process** button. This will transform the data, reference to TMS or the solvent, auto-phase the spectrum, and adjust the vertical scale. If you are happy with the results, you can skip to **Step 18** to print your data.



Note: This process runs automatically if you start the experiment with the **Go** button. Starting an experiment with ga will only transform (wft) the spectrum at the end of the experiment.

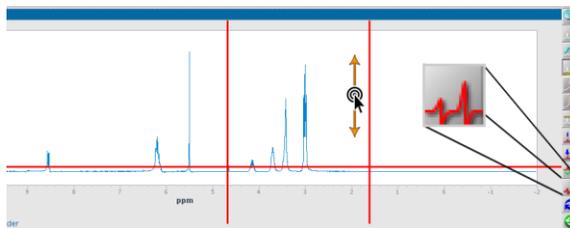
13. Basic Processing Tools

- A. Toggle between 1 or 2 cursors on the screen.
Also restores display mode.
 - B. Show full spectrum
 - C. Expand region (white space) to full window
 - D. Zoom in to the area between cursors. Set cursors by left-clicking on the left side of the region and right-clicking on the right side.
Then click this button to zoom in.
 - E. Zoom out
 - F. Click this button and then click-and-drag the region you want to expand/zoom in.
 - G. When zoomed-in, this tool can be use to move/scroll the spectrum left or right. It can also be used to adjust the vertical position of the spectrum.
 - H. (top) Display the time domain or FID.
Command: df
 - I. (top) Display frequency domain or spectrum.
Command: ds
- You can use the mouse scroll wheel (or click-and-drag with it) to adjust the vertical scale (height) of the peaks.
 - The **Weighting** page under the **Process** tab contains the tools to change the **line broadening** (lb), **transform size** (fn), and run **autophase** (aph0).



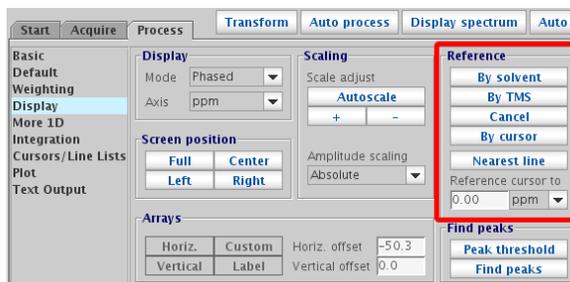
14. Phasing

To manually adjust the phase, click the phase mode button in the right tool menu. Then single-click a peak near the baseline on either the far right or far left. Click and drag vertically within the two red lines that appear to adjust the phase of the peaks within the region. Left click-and-drag will course-adjust the phasing and right click-and-drag will adjust more finely. If needed, click on a peak at the opposite end of the spectrum and repeat on that region to phase. Note that this second phasing step will have a larger affect on the whole spectrum and you might want to use the right mouse button to adjust. To exit phase mode, click the top button in the right tool menu (Step 13 A) or scroll within the spectrum window.



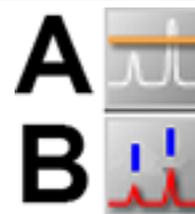
15. Referencing

- Zoom in on the reference peak and left-click near the center of the peak. Click the nearest line button in the right tool menu or enter `n1` on the command line.
 - Enter `r1 (#.##p)` on the command line where `###` is the desired chemical shift.
- The same tools can be found on the **Display** page in the **Process** tab. Click on the peak, click the **Nearest line** button, and then enter the reference value in the text box and press enter or click the **By cursor** button. The **By solvent** and **By TMS** buttons will auto-reference to the solvent peak or TMS peak respectively.

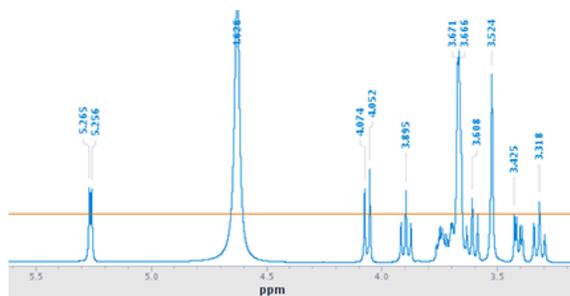


16. Peak Picking (Labeling)

- Click the display peak frequencies button in the right tool menu (**B**). Doing this first will allow the peak labels to dynamically update in the next step.

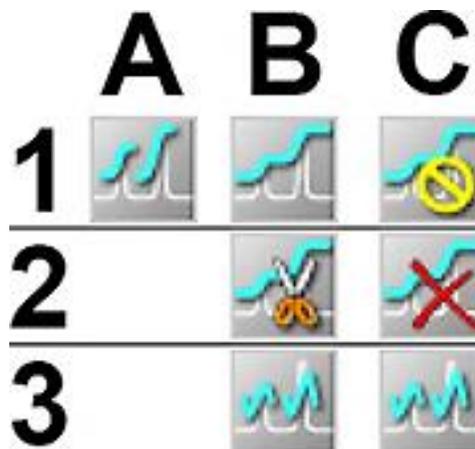


- Click the set threshold button (A). Vertically drag the orange horizontal line in the spectral window to adjust which peaks are labeled. To exit the set threshold mode, click button A again.



17. Integration

Integration in VnmrJ is a little different than other software packages. The toolbar changes for the integration buttons. When integral lines are turned off (normal mode), you will just see one button as in column A. Clicking a button in row 1 will cycle between columns A through C.



Turn off integral lines

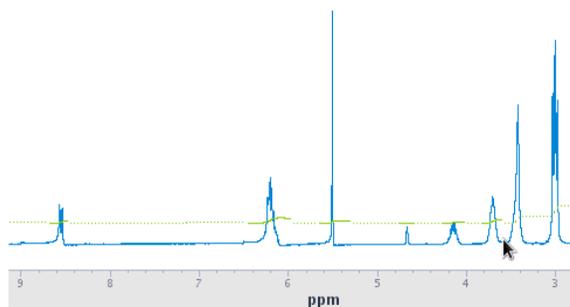
The peak vertical scale can't be adjusted while displaying the integral lines. To turn these off, click the yellow C1 or type `intoff` in the command line.

Delete all integral regions

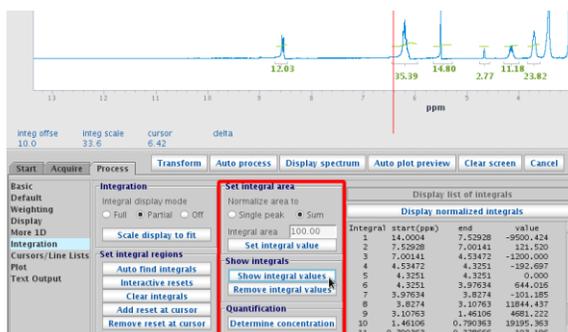
To reset/erase all integral regions, click C2 or enter `cz` in the command line.

Set integral regions

- Click B2 (orange scissors).
- Left-click to set an integral region point. It is best to click the left side of the peak and then click the right side of the peak to set the region. Defined regions (completed integrals) will show as a solid green line. The 'blank space' between integrals will show as a dotted line.
 - You can 'undo' or delete an integral region point by right-clicking near that location. This can be useful to expand/contract regions. You can delete the start or stop point of the region and then left-click at the new position.
 - To exit the set integral mode, click the display cursor button at the top of the tool menu (Step 13 A) or enter `ds` or `wft` in the command line

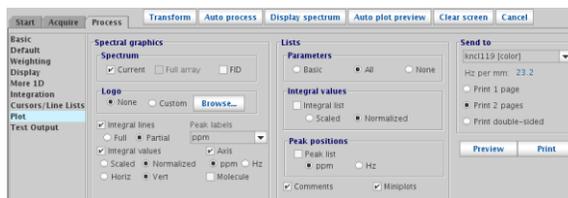


- Integral values can be displayed by clicking the **Show integral values** button on the **Integration** page under the **Process** tab. By default, this will display values whose sum is 100.
- To define the area of a peak, place the left red cursor on the peak, click the **Single peak** radio button, and enter a value in the **Integral area** text box on the **Integration** page and click the **Set integral value** button. Then redisplay the integrals.
- Use the **Sum** setting above to redefine the total integral from the default of 100.
- For accurate integrals, make sure the spectrum is properly phased and try to define the regions from baseline to baseline.



18. Print Spectrum

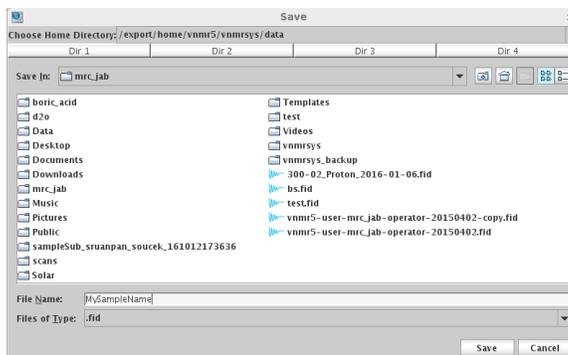
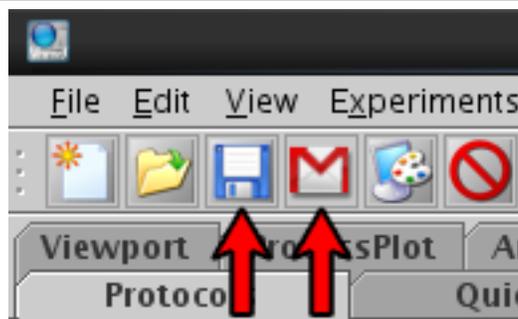
All basic print options can be found on the **Plot** page under the **Process** tab. Toggle the options you want and click **Print**. The **Preview** button can be used to preview the selected options or to save a PDF of the spectrum instead of printing.



19. Saving and/or emailing data

Save

- **Data is not automatically saved on the walkup instruments! You must save/email the data after each experiment or the data will be lost.**
- Click the Save As floppy icon or **File>Save As...**
 - Navigate to the desired directory, enter a file name, and click Save in the popup window.
- It is recommended to verify the file has saved by clicking Save As again and checking the file is listed in the directory.



Email (send data as a .zip file)

- Click the Gmail icon or **File>Email the FID**

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- If a sample name and/or email was not provided before running the experiment, you will be prompted on the command line.
 - The email address must be either an @zips.uakron.edu or @uakron.edu address.



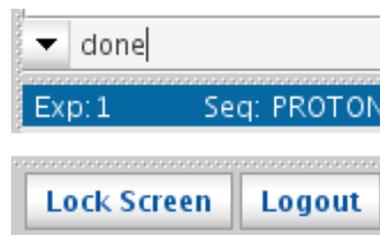
▼ Enter a sample name: MySampleName|
▼ Enter your email address: nmr123@uakron.edu|
Exp: 1 Seq: PROTON Index: 1

20. Continue Experiments or Samples

- Goto Step 9 to continue running experiments on the same sample
- OR**
- Goto Step 2 to continue with your next sample

21. Finish

- Click the **Start** tab
- Check the sample height of the standard (D₂O) sample using the depth gauge.
- Replace your sample with the standard.
- Click **Logout** in the **Start** tab or enter done on the command line
- Complete the logbook with your logout time and any missing information.



▼ done|
Exp: 1 Seq: PROTON
Lock Screen **Logout**

ALWAYS VERIFY THAT YOU HAVE LOGGED OUT BEFORE LEAVING!

You will be charged based on the amount of time you are logged onto the instrument. To avoid excess fees, make sure you see the login screen in Step 1 before you leave.