NMR New User Workshop

Second session

Rui Huang

Notes reproduced from Dr. Weiguo Hu

Link:

ruihuang@umass.edu

weiguoh@polysci.umass.edu

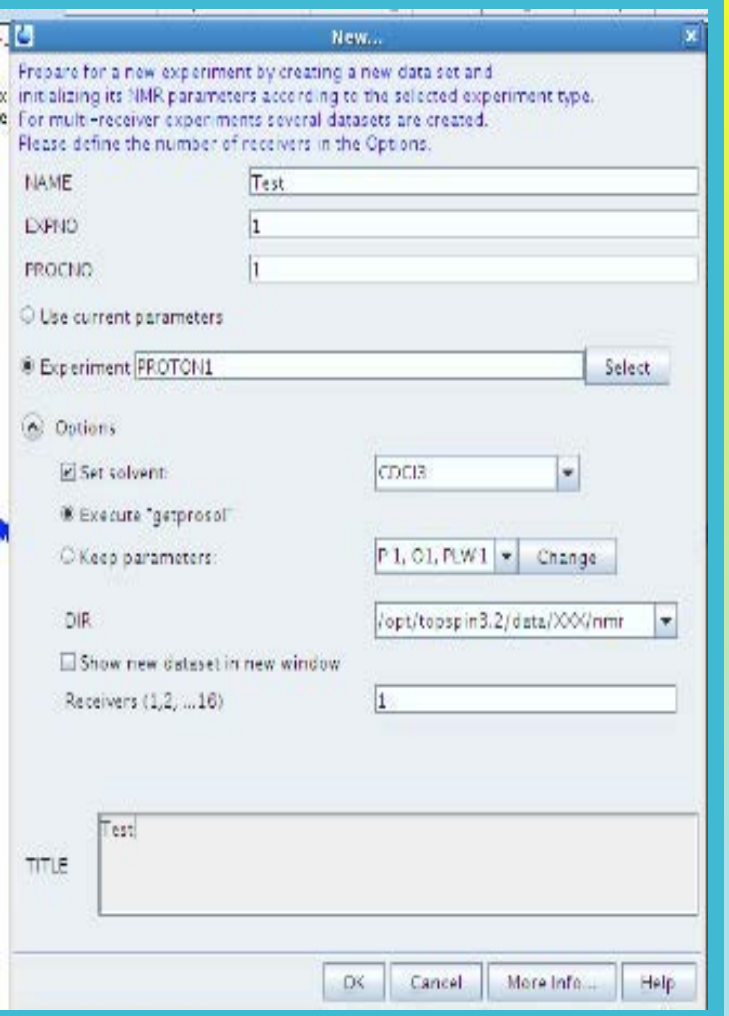
Booklet:

https://udrive.oit.umass.edu/weiguoh/nmrhandouts/Introduction-NMR.pdf

Questions and Hands-On Practices

- What are some safety concerns in the NMR lab?
- What are some best practices when preparing NMR tubes for experiments?
- What are the purposes of atma, lock, shim, and rga?
- What are some necessary spectral processing steps to take before integrating the peaks?

^{13}C NMR



- edc and select CARBON1 in experiment (attach the screenshot)
- *atma*
- Sensitivity of ^{13}C: ^{1}H
  - The more sample amount, the better the s/n will be
  - Using line broadening (lb) to enhance s/n
- Every carbon has one single peak
  - Because a "decoupling" technique is used to remove influence from 1H
- Integration is less reliable than 1H

^{13}C integration can be very useful if used wisely

https://blogs.umass.edu/weiguoh/

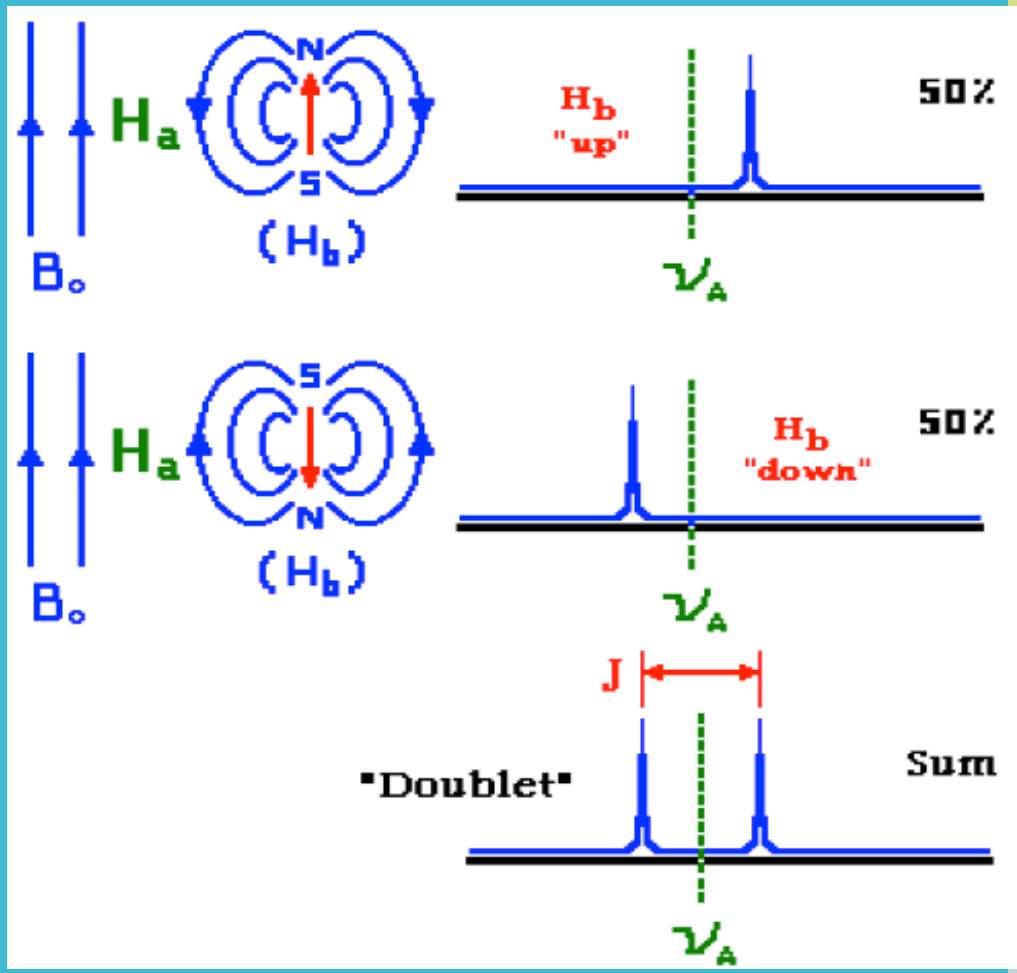
Decoupling

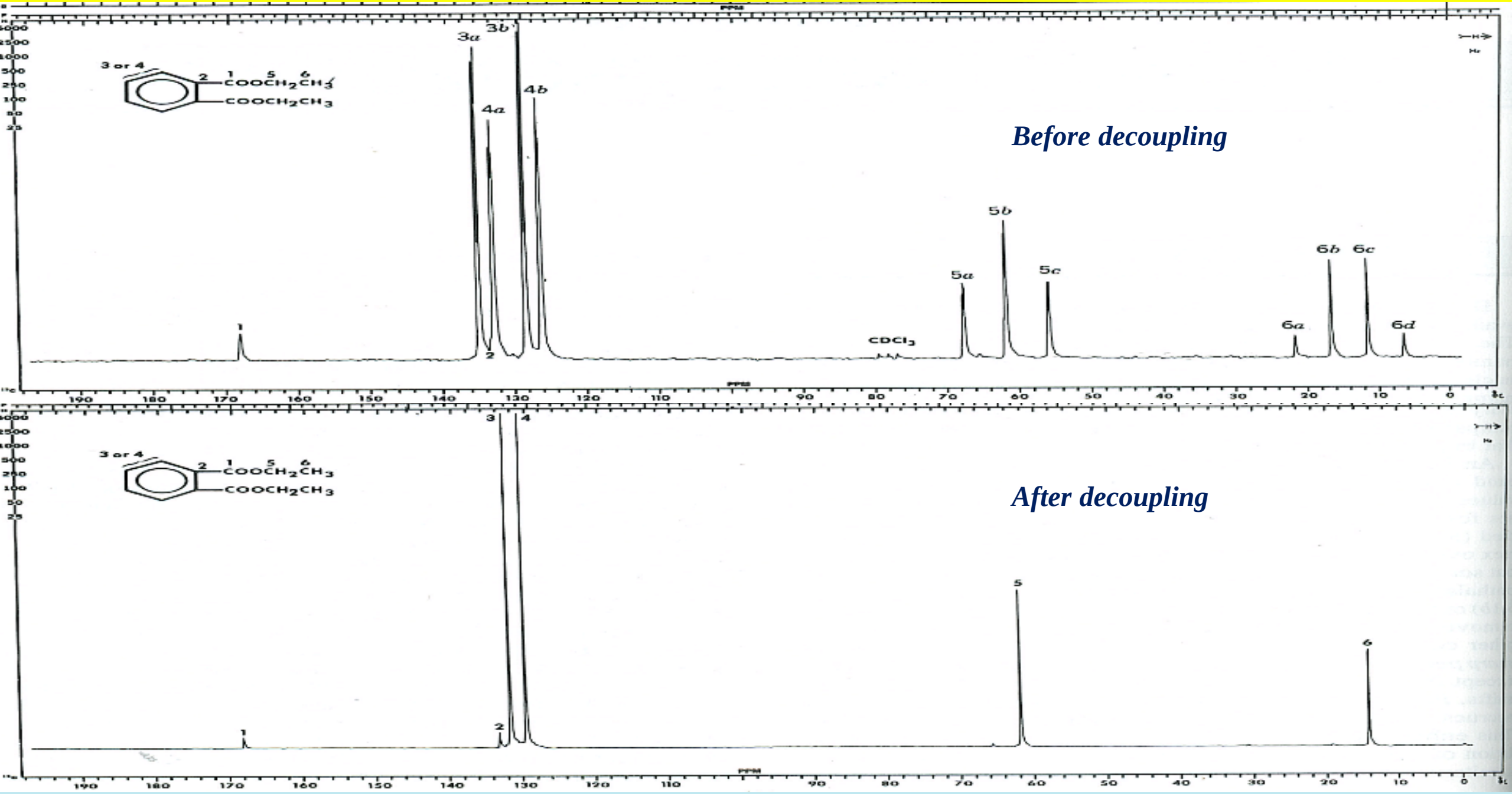
In ^{13}C experiment, ^{13}C-^{1}H J-coupling is usually removed by a "decoupling" pulse sequence

– Pulsing at ^{1}H during detection of ^{13}C signal
- i.e. Set pulsing frequency to ^{1}H and detector frequency to ^{13}C

– Decoupling resolves overcrowding due to large ^{1}J splitting
- The large ^{1}J couplings usually don't provide useful information

– Decoupling reduces ^{13}C multiplets to singlets, which improves S/N

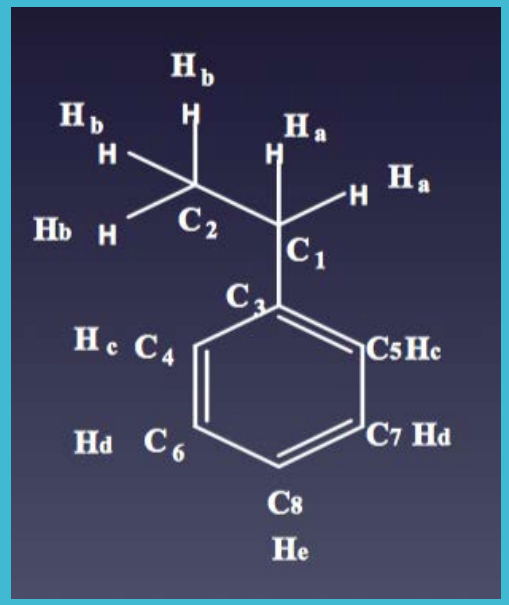


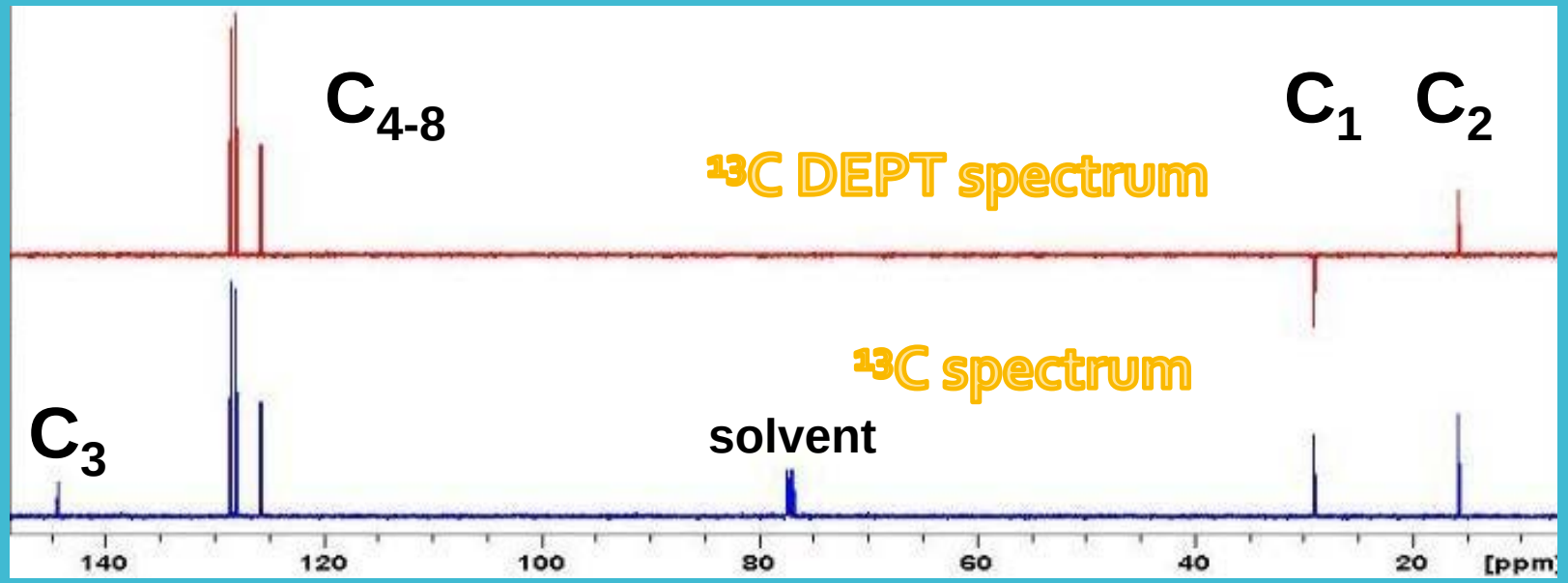

3 or 4
COOCH2CH3
COOCH2CH3
3a
3b
4a
4b
1
2
CDCl3
5a
5b
5c
6a
6b
6c
6d
Before decoupling
3 or 4
COOCH2CH3
COOCH2CH3
3
4
1
2
5
6
After decoupling

DEPT (Distortionless Enhancement by Polarization Transfer)

Discern methyl, methylene, methine, and quaternary carbons

- **A ^{1}D technique, with several variations. Most popular is DEPT-135**
- **CH and CH3 peaks are positive; CH2 peaks are negative**
- **Carbons with no directly bonded protons do not have signals**
- ***Why do solvent peaks vanish in DEPT?***





2D NMR : implementation

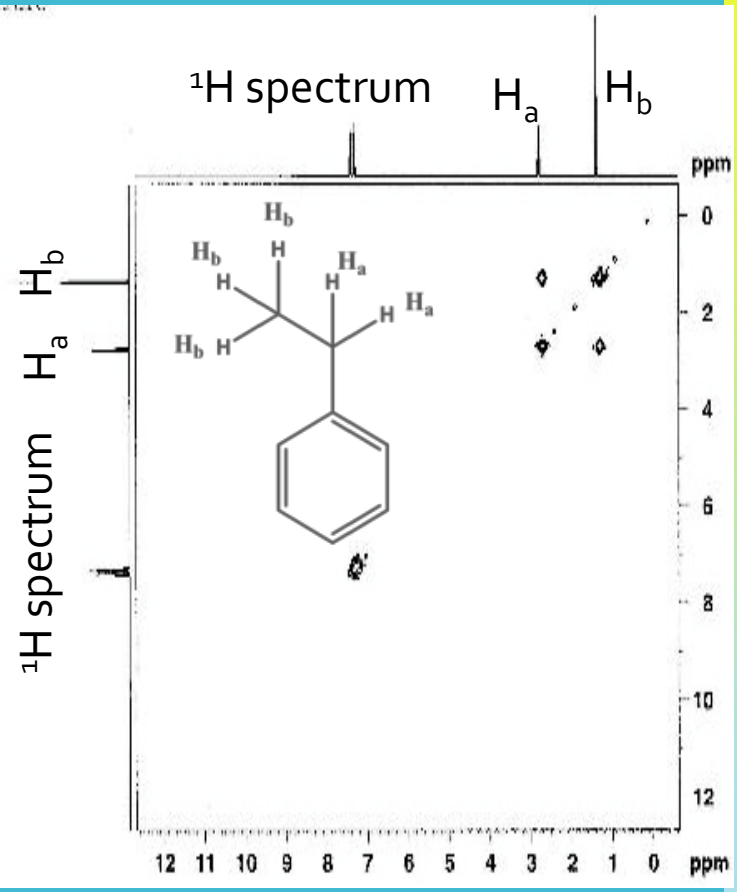
•Conventional NMR spectra (one-dimensional spectra) are plots of intensity vs. frequency

• In two-dimensional spectroscopy intensity is plotted as a function of two frequencies.

COSY : ^{1}H and ^{1}H correlation : *Connectivity of protons*
HMQC: ^{1}H and ^{13}C correlation : *Direct C – H Connectivity*
• edc to create the directory
• Running a 2D experiment is almost as easy as running a 1D

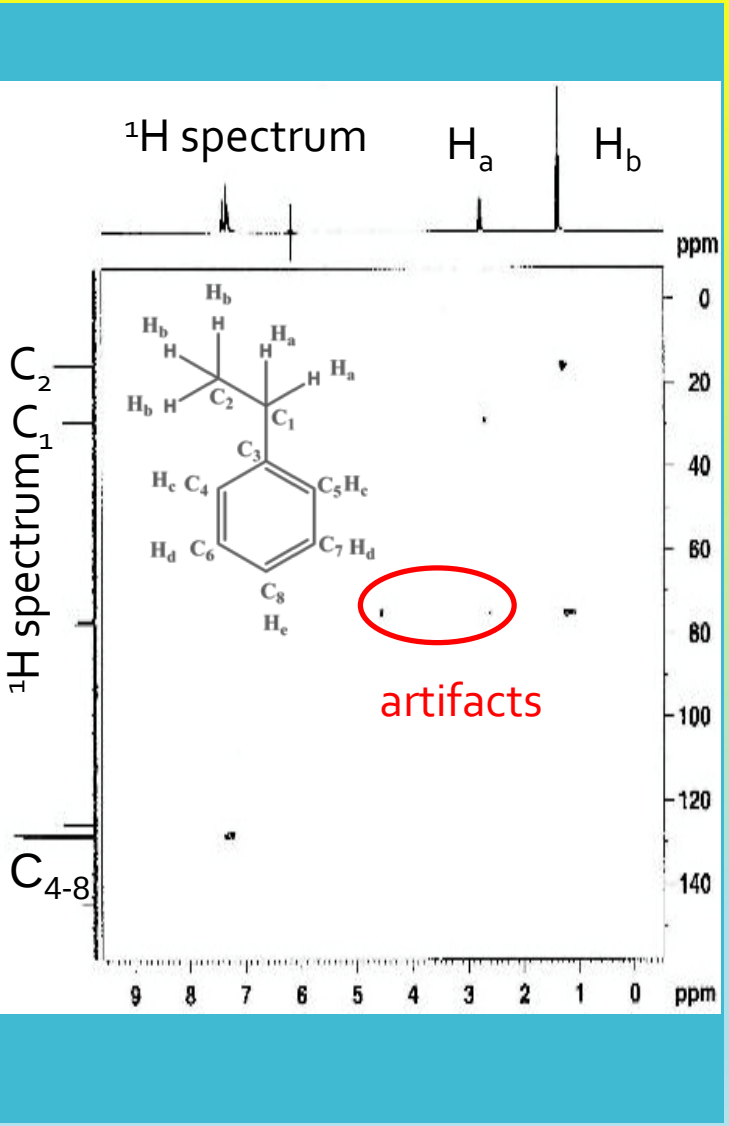
– All the pulse sequence parameters have been “packaged” for you

COSY(Correlation Spectroscopy)



- Detects connectivity between protons that have J coupling
- • Both dimensions are ^{1}H
- • Vertical is always 1st dim.
- • 3 <u>diagonal peaks</u>
  - – provide no new information
- • <u>Cross peaks</u> at (1.2ppm, 2.6ppm) and (2.6ppm, 1.2ppm) indicate strong J-coupling
  - – Usually only 2 J and 3 J are strong enough to give cross peaks
- • You can optionally plot 1D spectrum on the side to compare with the 2D spectrum. 1D spectrum usually has higher resolution and less artifacts.
- • **Fourier transform command:** *xfb* instead of *efp*
- – *xfb* be can done at any point during the analysis
- – when good resolution is achieved the experiment can be stopped
- using *halt* command

HMQC (Heteronuclear Multi-Quantum Correlation)



A ^{13}C-^{1}H 2D spectrum

- No diagonal peaks

  -Diagonal peaks could only exist when both dimensions are the same type of nuclei

- Shows direct C-H connectivity
- Non-protonated carbons don't have a peak on the spectrum
- Beware of artifacts

  -Along the "ridges" of big peaks

  -In the middle lines of spectrum window

- Must do *atma*

NMR Facility Policies

1.Reservation

–please follow the reservation rules on top of the Calcium calendars.

–3 violations will face suspension of their accounts for 3 weeks.

2.Internet access is limited to umass domain

3.Data storage: The NMR computer gets full quickly. You need to transfer out data and delete them from the NMR computer asap.

–pse server automatically deletes data that are > 7 days old

–Don’t use flash drive; it could contaminate

–If you have too many data files, Tospin will load very slowly

–You have two folders to store your data:

/home/[username]. **This includes your desktop. Intended for temporary storage only.**

/opt/topspin3.2/data/[username]/nmr. **You can keep stuff here for slightly longer.**

NMR Facility Policies

5.Reporting of problems and incidents

–Report instrument problems to Czar if your group has one, otherwise report to me.
–Never attempt to fix problem or reboot computer
–Report incident caused by you (standard sample breakage etc.) to Dr.Hu

6.You are responsible for the act of anybody using your account (junior grad students, summer students et al)

–When you feel your trainees can comfortably run simple NMR experiments themselves, ask them to contact me to obtain their own accounts

7.Please keep the workplace clean