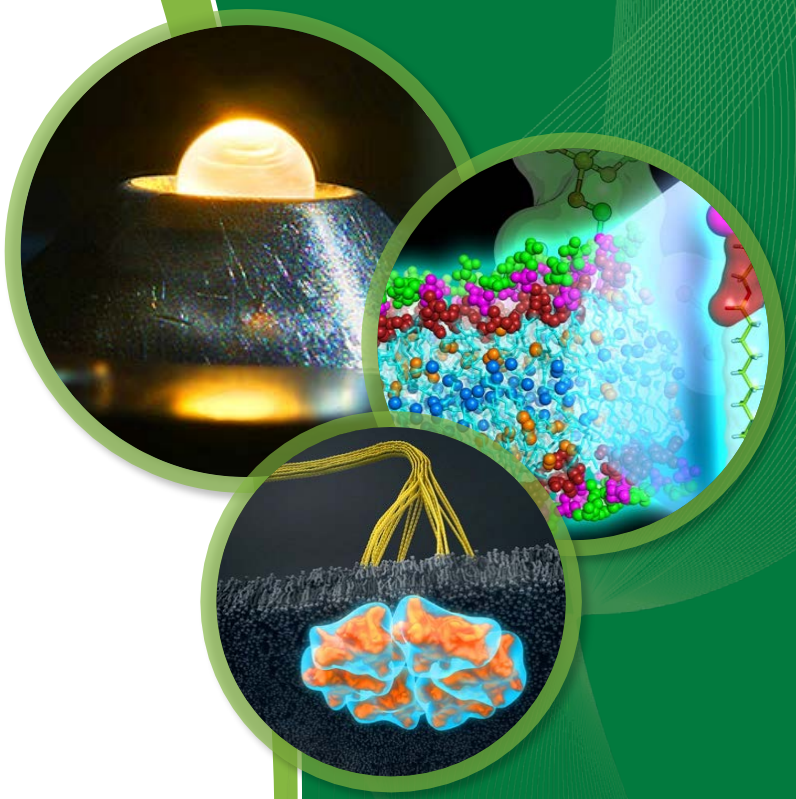
Dynamic Nuclear Polarization for Neutron Scattering



Josh Pierce

Neutron Technologies Division

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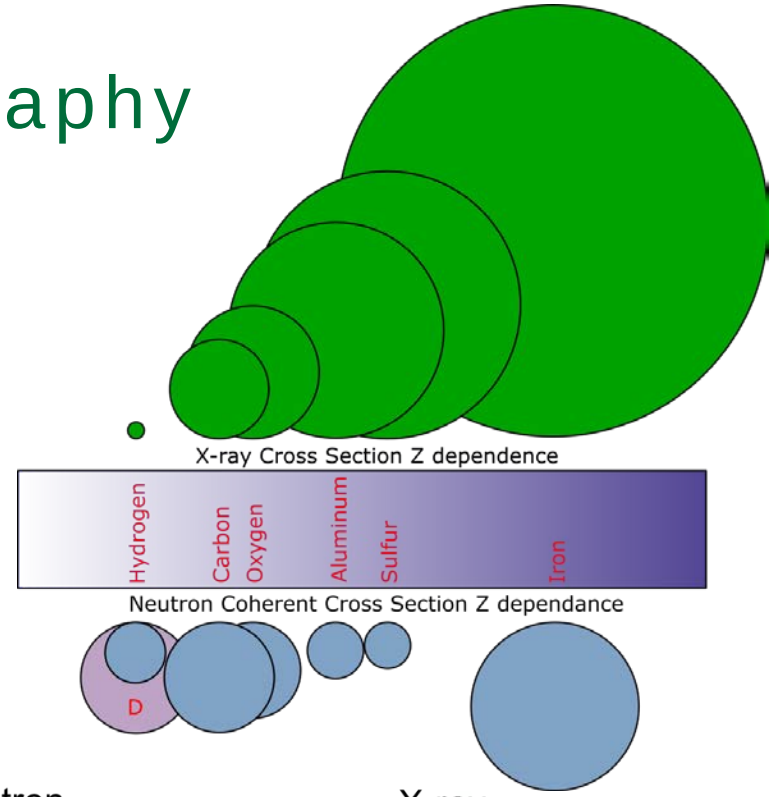
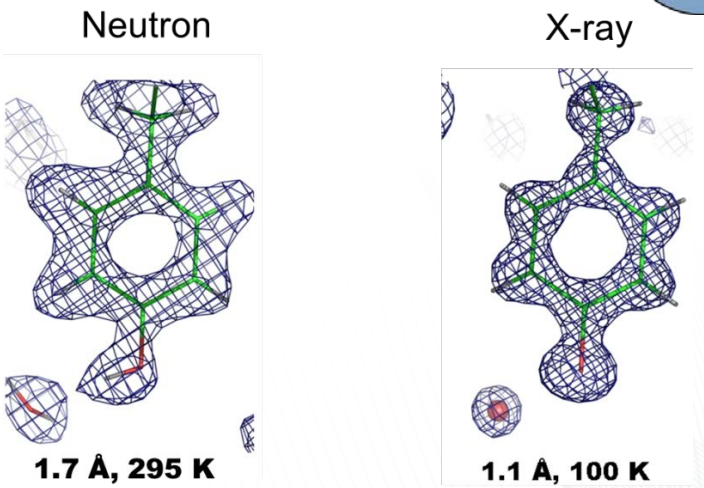
Neutron protein crystallography

- Usually, macromolecular crystallography uses X-ray facilities to measure molecular structure
  - Modern light sources have incredibly high flux
- Using neutrons for crystallography has pros/cons:
  - Comparatively low flux
  - Sensitivity of the neutron cross section to lighter elements (especially hydrogen)
  - Sensitivity to isotopes
- NPX is a unique experimental tool for the experimental location of key hydrogen atoms and water molecules in biological macromolecules.





Gardberg *et al.* (2010) *Acta Cryst.* D**66**:558-567

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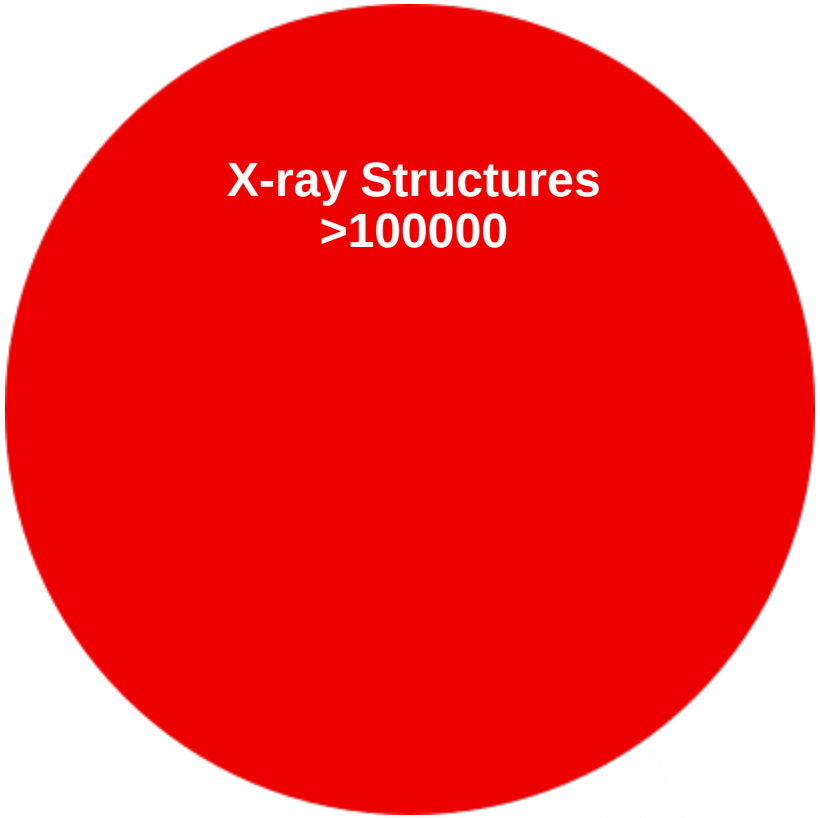

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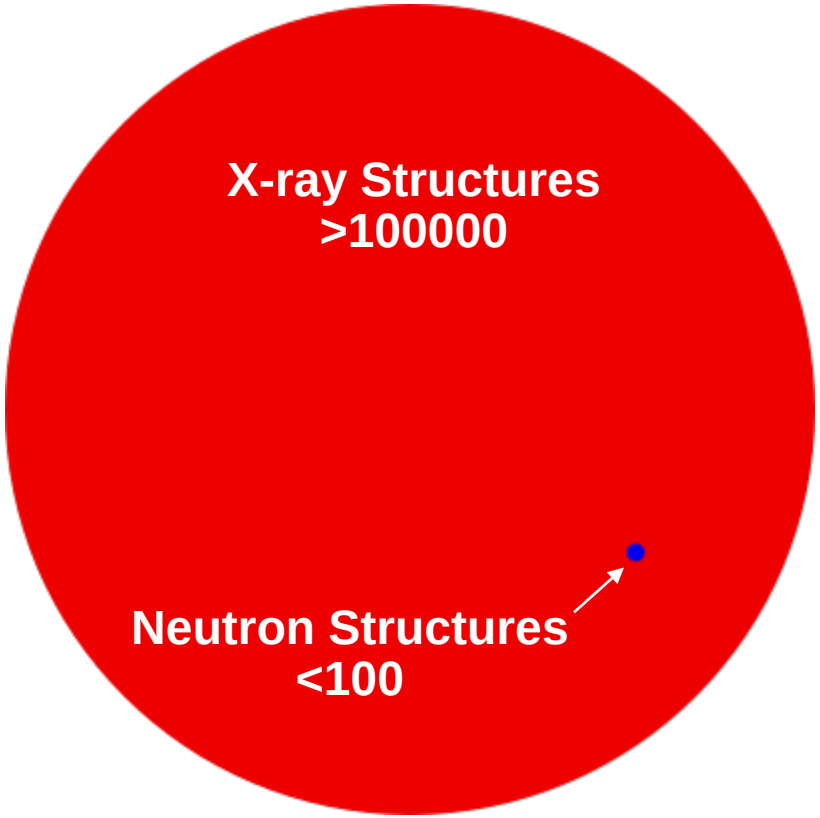
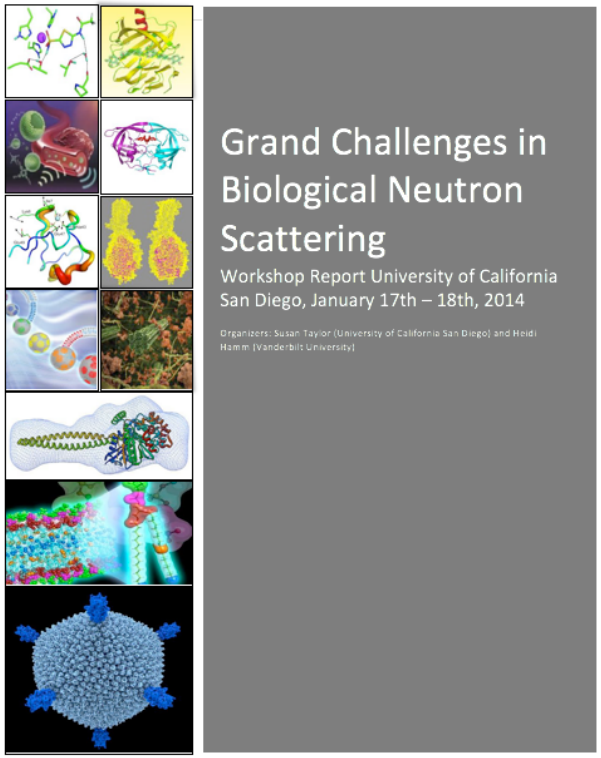


X-ray Crystals <0.001mm^3
Neutron Crystals >0.1mm^3

Community Scientific Needs: Smaller Samples and Faster Data Collection



At the workshop, 37 invited leading researchers from more than 20 different universities and institutes joined 5 participants from the Neutron Sciences Directorate of ORNL to map out 10 grand challenges that we face in biological research over the next 10 years.

- **Grand Challenges: Cold neutron flux:** **"**Radically increase the flux of neutron beam lines at long wavelengths in particular for small angle scattering, crystallography, and spin echo."

Gains Required

- Data set in 1 day x20
- Reduce crystal size to 0.001mm^3 x100

- How can we accomplish this without building a new facility?

Spin Dependence of Neutron Scattering

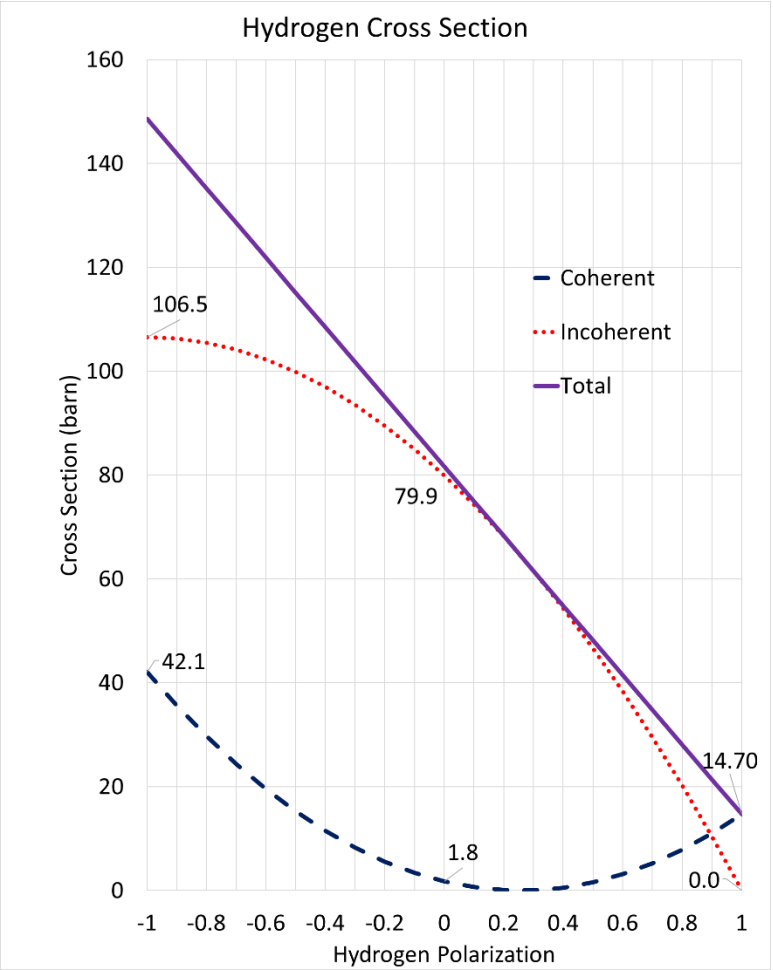
- For a lattice of identical atoms with non-zero spin, the incoherent and coherent cross section for neutron scattering has a dependence on the spin alignment of the neutron and the struck nucleus
- Control over spin orientation gives control over scattering.
- Neutron Polarization is well developed
  - Supermirror polarizers
  - 3He filters
- Nuclear Polarization is more challenging

$$\left(\frac{d\sigma}{d\Omega}\right)_{inc} = \frac{b^2}{4}\{I(I+1) - pPI - P^2I^2\};$$

$$\left(\frac{d\sigma}{d\Omega}\right)_{coh} = \left\{b_0^2 + bb_0IpP + \frac{I^2P^2b^2}{4}\right\};$$

Spin Dependence of Neutron Scattering from Hydrogen

- Hydrogen is a special case
  - The spin dependence of the hydrogen cross section is very large
  - Looking for hydrogen locations is a primary motivation for Neutron Protein Crystallography
- Incoherent scattering can be removed entirely (true for any nucleus)
- Coherent scattering can be increased by a factor of 7 (or 20)
- An increase in signal to noise enters squared into the calculation figure of merit
  - Factor of 10 in signal to noise is a factor of 100 in flux/sample size/data collection time
- The hydrogen nucleus is polarizable



Coherent, incoherent and total scattering cross section of hydrogen as a function of the proton polarization for fully polarized neutrons.

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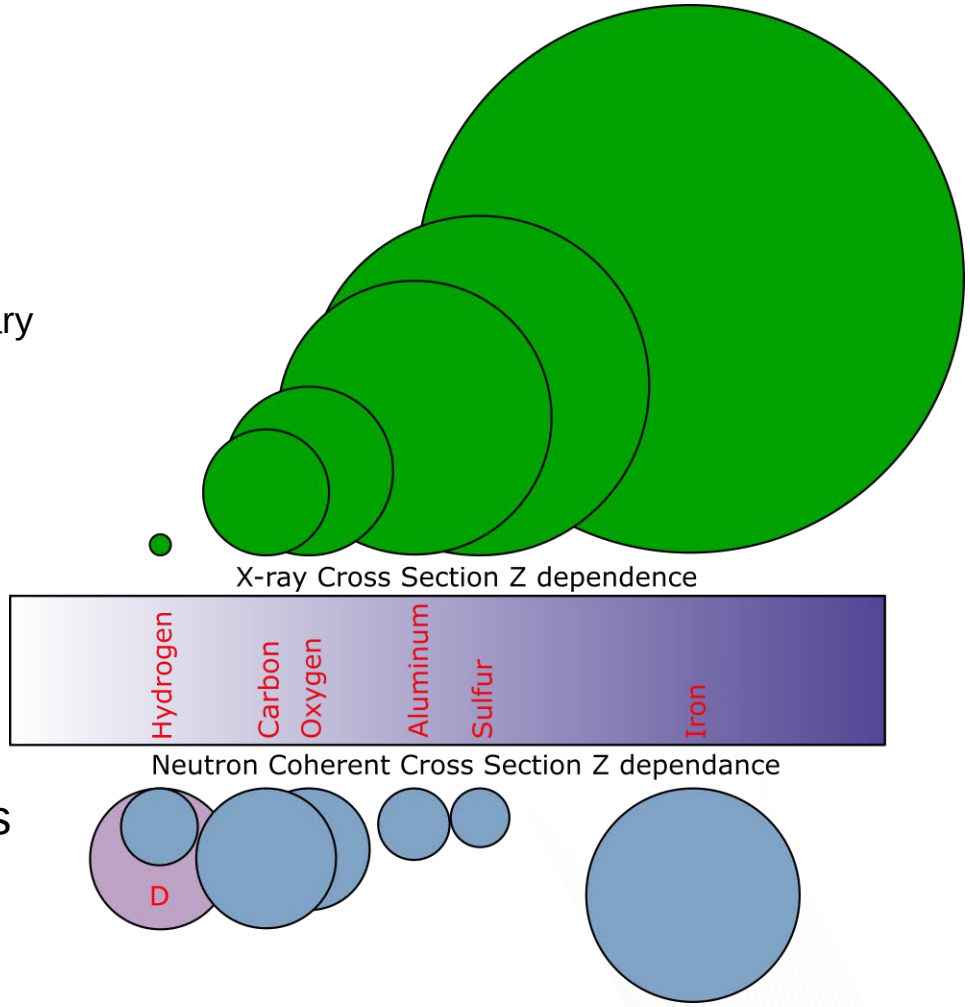

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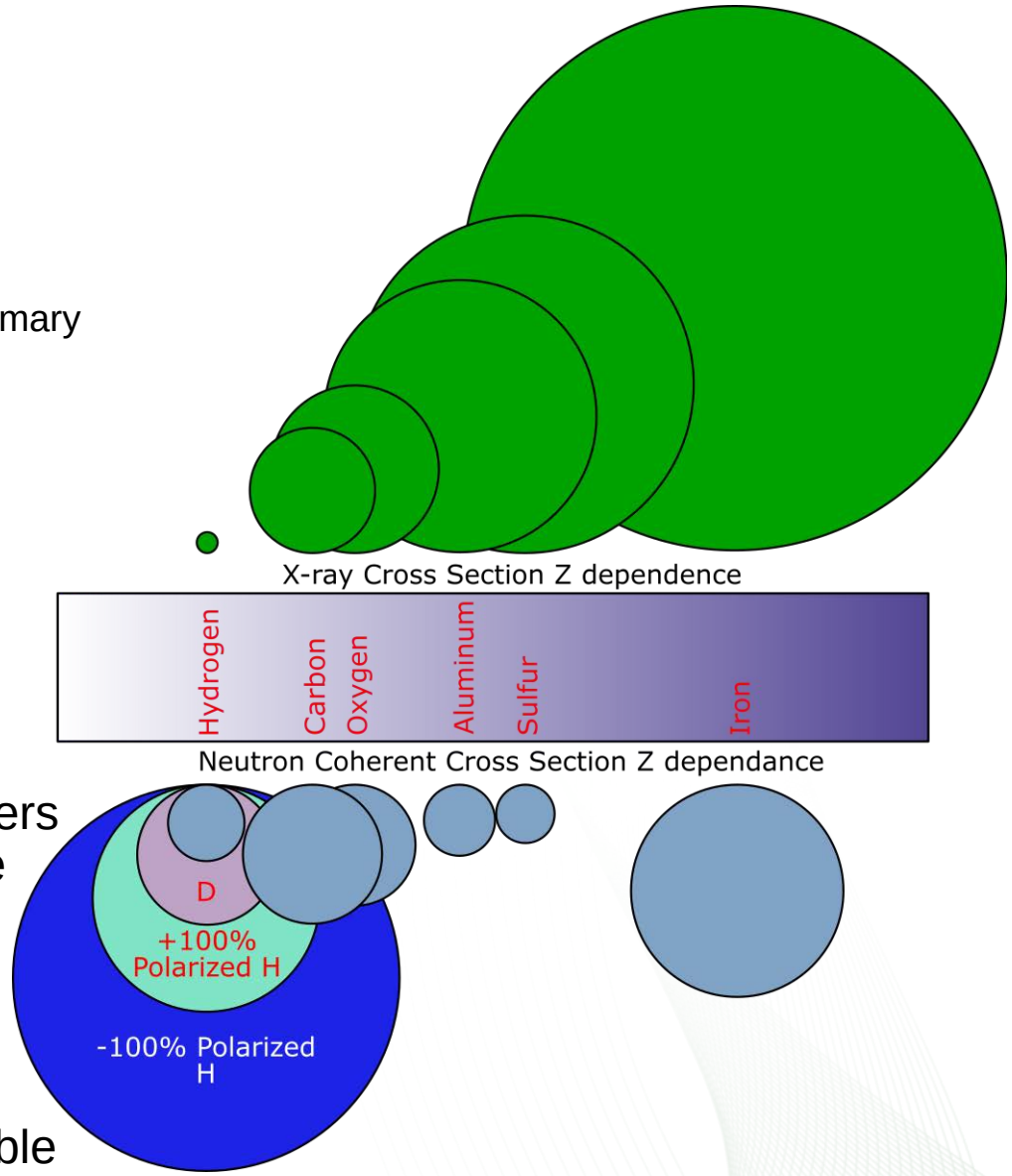
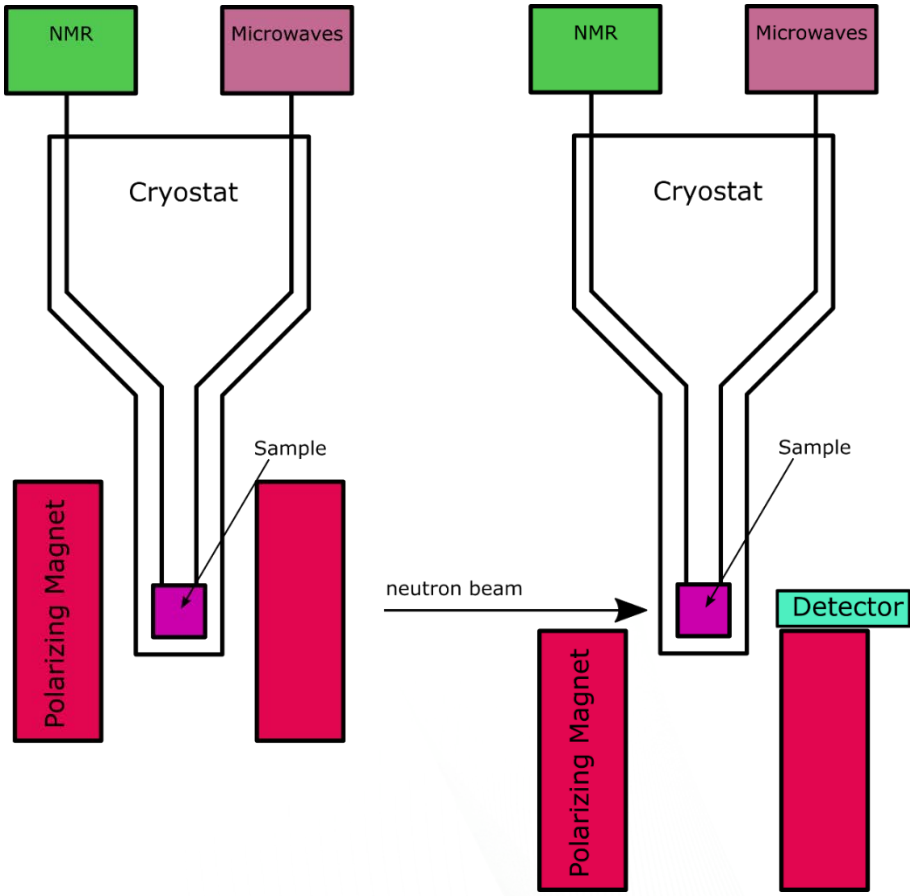


Tests at IMAGINE

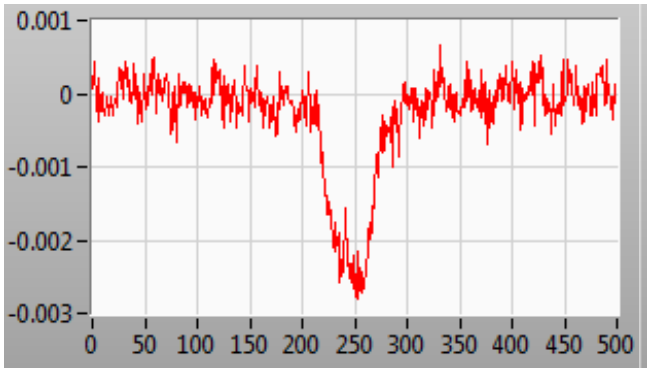


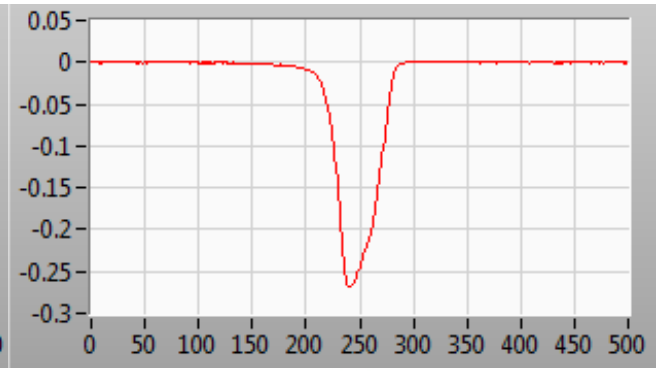
Refrigerator and Magnet

- Stock Dilution Refrigerator from Bluefors
  - 400μW@100mK
  - >1mW@1K
  - Base temperature (unmodified) ~7mK
  - Cryogen Free
- 5T Solenoid
  - Warm 100mm bore
  - Cryogen Free

NMR

- Absolute polarization measurements are challenging
  - Target sizes are generally <1mm^3
  - TE signals are extremely small
- Liverpool Q-meter *almost* works
  - With sufficient gain and careful work, a decent TE can be measured
  - Error is high
  - Still hard to trust
    - Sensitive to very small contaminations

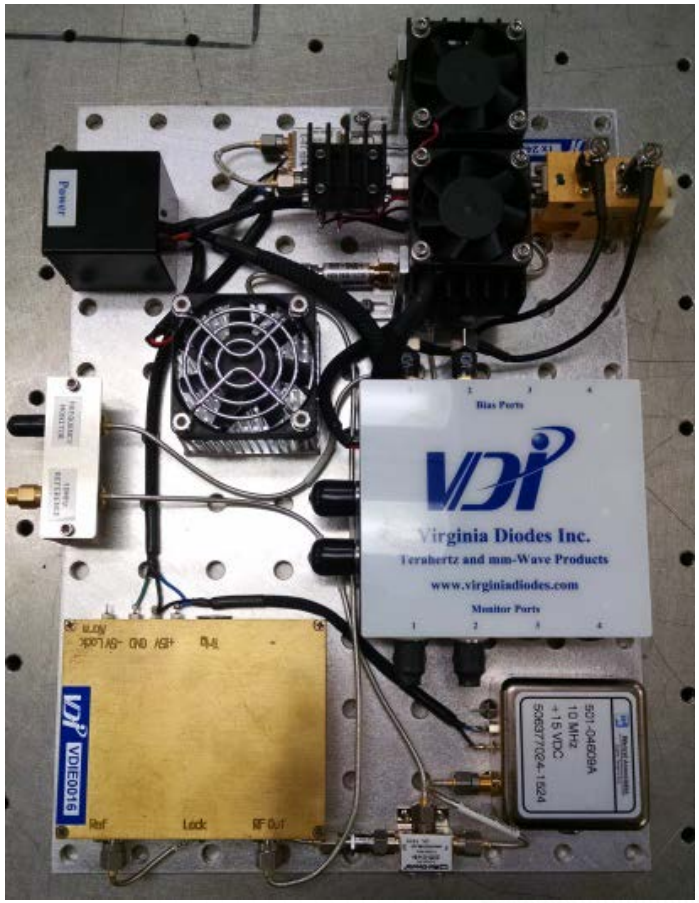




- In general, absolute polarization measurement is not needed
  - The higher the better
  - Absolute numbers may be extracted from diffraction data
  - Large solution samples are used to test performance

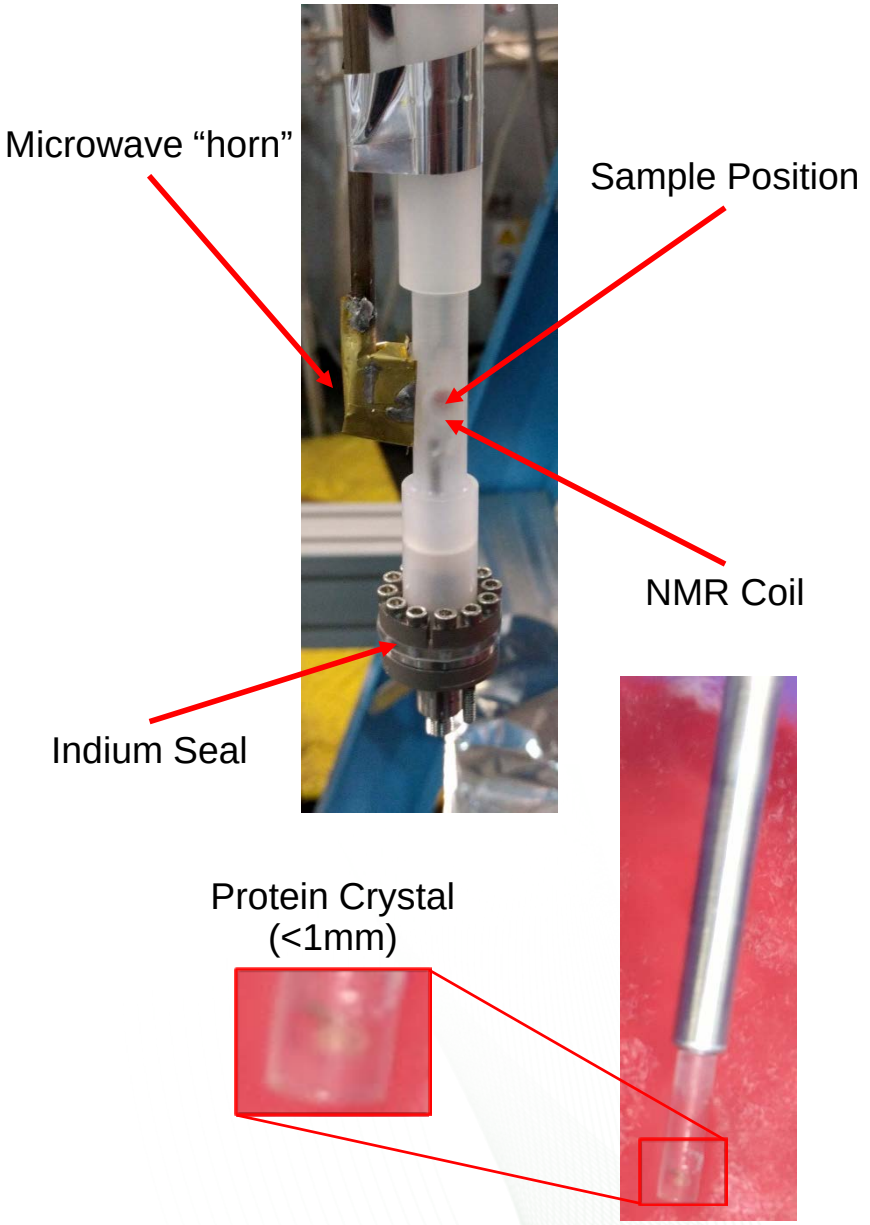
Microwave System

- Here, small sample size is helpful
  - Very low microwave power needed because the sample is tiny
- VDI diode based microwave source
  - Low cost
  - 300mW of power @140GHz
  - Ease of use
    - USB control
    - Incredibly stable frequency
    - No high voltage or cooling



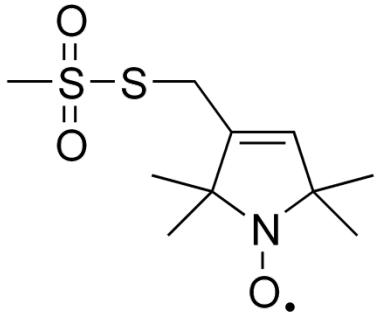
Sample Interface

- Requirements
  - Cannot put the sample in the mixing chamber
    - 3He captures neutrons
  - Allow crystal rotation
  - Fit in 100mm space
  - Load sample cold
    - Crystals cannot warm past ~150K
- Sample located in a Kel-F chamber, surrounded by superfluid
  - Not sealed space
  - Large cryogenic load from 4He film/vapor
  - Hard to get repeatable temperatures
    - Sample temperatures vary from 300mK to 150mK

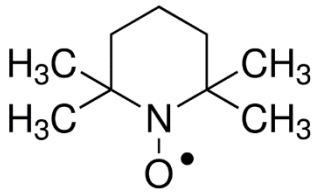


Paramagnetic Labeling in Protein Crystals

- Strategies
  - Site specific
  - Non-specific
- Site specific labeling (Intrinsic)
  - Mutagenesis for intrinsic/site specific labels
  - Spin-labeled mutants T4 lysozyme constructed, expressed, crystallized
  - X-ray and first neutron structures determined
- Non-specific
  - Crystals soaked in a solution with paramagnetic label



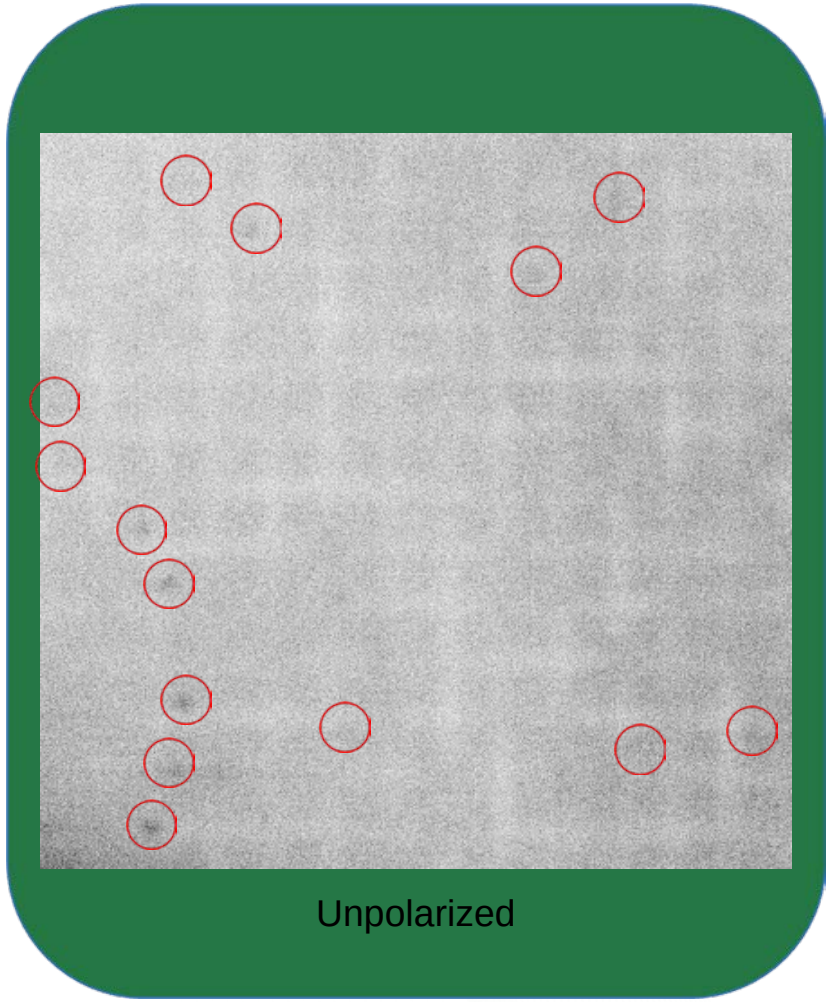
MTSL



TEMPO

T4 Lysozyme Results

- Doped with TEMPO
- “Large” crystals
  - ~0.5mm-1.0mm on edge
- Detector was uncalibrated, and shifted between frames
- Short hold times in “frozen spin” mode
  - ~60-180 min T_1
  - Very high temperatures
    - ~230 mK
- Measured diffraction pattern change
- Enhancements of 2-3 in integrated diffraction pattern for anti-aligned spins
  - The enhancement of individual reflections depends varies depending on the relative contribution of hydrogen
- Consistent with maximum polarizations of around 50%



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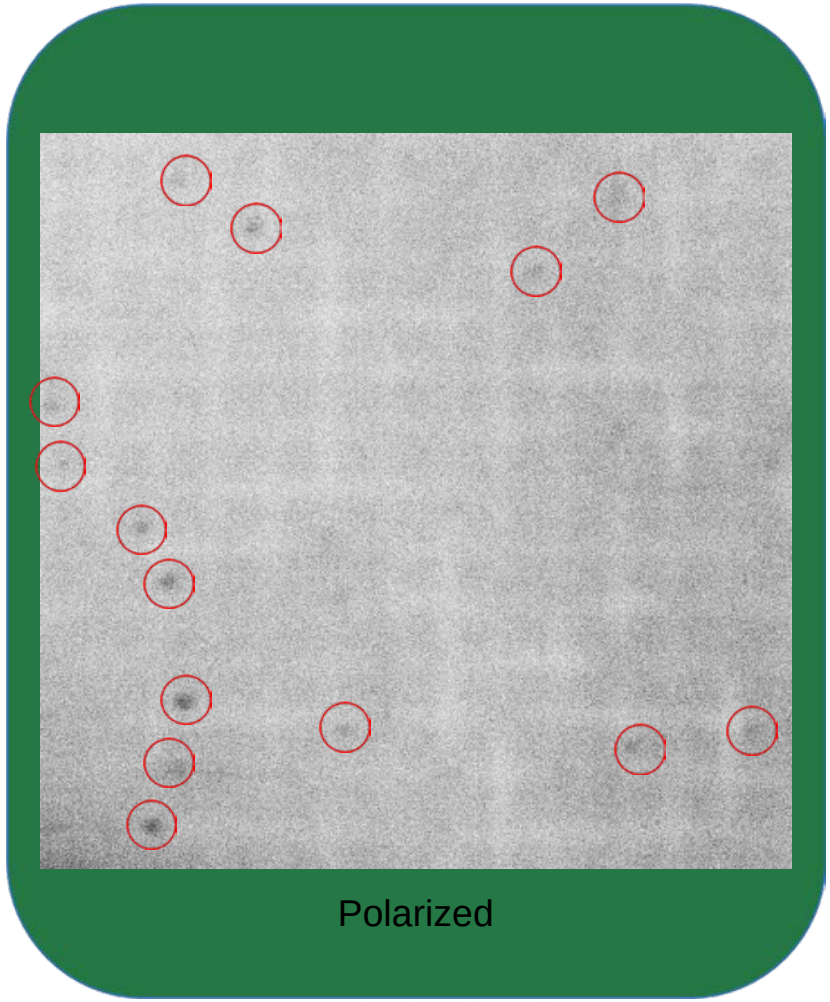


Moving Forward

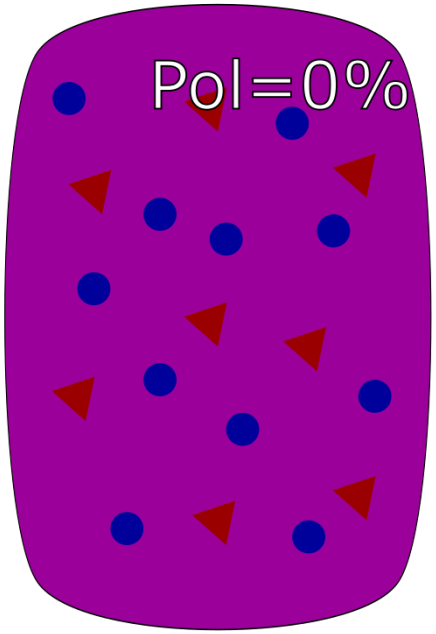
- New Scattering data will be taken starting in November 2018
- System has been updated with ¼” VCR based cold seal at the top of the sample stick
  - Seal made at low temperature using 0.005” KELF gasket
    - Appears to be very reliable and require little torque
  - Relaxation times improved by factor of 1.5-2.2
- Improved (existent) DAQ software
  - Will allow the crystal orientation to be determined
  - Allow extraction of polarization from scattering data
- New detector
  - Same design concept, but lower noise

Conclusion

- The nuclear spin dependence of neutron scattering can be used to manipulate scattering
- Polarized hydrogen has a large potential benefit to protein crystallography
  - Could allow the use of sustainably smaller samples or greatly reduced data collection time
- DNP is an effective means to polarize the hydrogen within a sample
  - Sample preparation and sample environment requirements are substantial

Application to SANS Contrast Matching

- Deuteration
  - The unpolarized coherent scattering length of hydrogen is -3.74 fm
  - The unpolarized coherent scattering length of deuterium is 6.674 fm



- Polarization
  - Positive polarization: 10.82 fm
  - Negative polarization: -18.3 fm
  - Can be changed in-situ
    - Requires a single sample reparation
  - Work done by Stuhrmann et al
  - Current work by Kumada et al. (also working on DNP for reflectometry)

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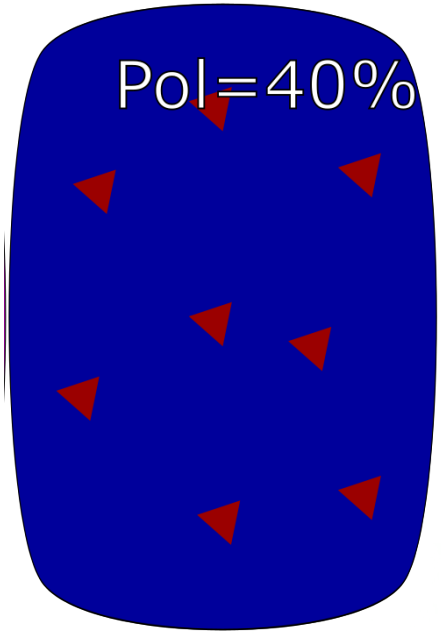

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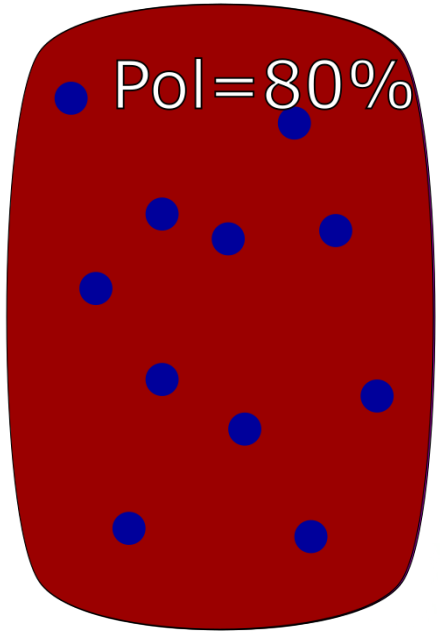

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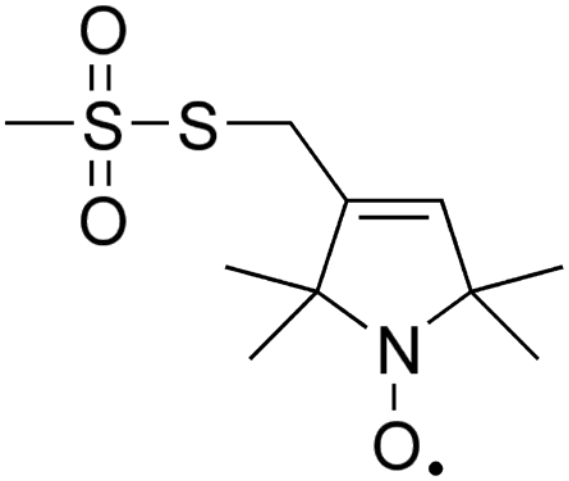
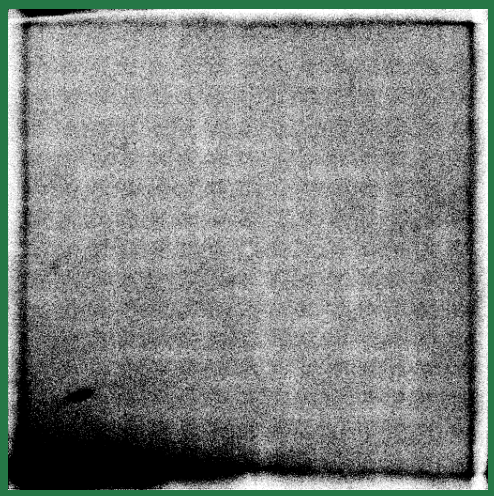
Advanced Techniques: Localized Polarization

- If the center is an attached spin label, then the location of the polarized region can be controlled
  - Attached to a specific site on a macromolecule
  - Size and rate of propagation has been studied with SANS (van den Brandt 2006)
- Alternatively, different components of a composite sample could be selectively polarized
  - One layer of a sample for example

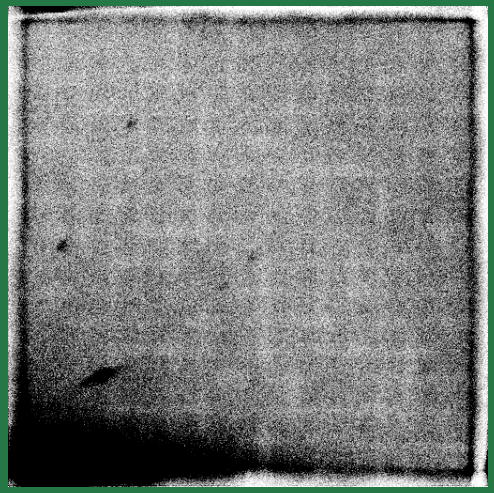


Advanced Techniques: Difference Measurements

- All that is required is to change the microwave frequency to change polarization sign
  - Field remains constant
  - Temperature remains constant
- Adiabatic Fast Passage or neutron spin flipper can reverse polarization more quickly
- Only thing that changes is the cross section for the nuclei, and that changes in a predictable manner
- This can be used to highlight specific structures



Spins Aligned



Spins Anti-aligned

Other Nuclei

- All non spin zero nuclei will polarize
- Polarization will be different for different nuclei
  - 15N and 13C polarize well, are used for DNP enhanced MRI and NMR measurements

$$a = b_0 + b\boldsymbol{I} \cdot \boldsymbol{s}$$

$$b = \frac{2(b_+ - b_-)}{(2I+1)}$$

- Spin dependent scattering lengths are different for each nucleus
  - 15N and 13C have very little spin dependence

$$\left(\frac{d\sigma}{d\Omega}\right)_{inc} = \frac{b^2}{4}\{I(I+1) - pPI - P^2I^2\};$$

$$\left(\frac{d\sigma}{d\Omega}\right)_{coh} = \left\{b_0^2 + bb_0IpP + \frac{I^2P^2b^2}{4}\right\};$$

- Difference measurements may still be possible
  - Different nuclei species could be selectively polarized/depolarized/flipped using a combination of NMR and DNP techniques