Symmetry and Spin: Improving Molecular Imaging with Molecular Hyperpolarization

Warren S. Warren, Duke University
Departments of Chemistry, Physics, Biomedical Engineering and Radiology
Director, Center for Molecular and Biomolecular Imaging

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1895: X-rays first image opaque media

Hand mit Ringen: print of Wilhelm Röntgen's first "medical" x-ray, of his wife's hand, taken 22 Dec 1895

An X-ray picture (radiograph) taken by Röntgen of Albert von Kölliker's hand at a public lecture 23 Jan 1896

(X-ray) Computed Tomography gives beautiful images of structure, but…

X-ray contrast comes from larger scattering cross section of heavier nuclei.

Contrast agents: Ba, I, Gd, … But does this correlate with disease?

Nuclear magnetic resonance (NMR) is very useful for studying molecules

Basic physics (1920s): atoms have “magnetic dipoles”, meaning they act like bar magnets (or spinning charged balls). Commonly called “spin angular momentum”

Basic physics (1940s): hydrogen atoms in a magnetic field have two stable states—“spin up” (pointed along the field, like a compass needle) or “spin down”

1960-present: chemistry, materials science, clinical diagnosis, oil well logging, watching thought . . .
Magnetic resonance imaging uses gradients to give high-resolution maps.

- **Gradient off**
- **Gradient on (0.01 Tesla/meter)**
- **Irradiate at one frequency**

![MRI Image with labeled anatomical structures]

- Pituitary Gland
- Sinus cavity
- Internal Carotid Artery
- Temporal Lobe
- Basilar Artery
- Pons
- Cerebellum
- Occipital Lobe
- Sulcus
Magnetic Resonance Imaging-the fundamental challenges

- Weak signal: $\approx 10^{-5}$ fractional magnetization
  
  *Detecting metabolites hard (particularly unlabeled)*
  
  *Imaging anything but water difficult*

- Water images structure more than function
  
  *Density, relaxation rates, diffusion: physics of 1954*

- Tissue-the ultimate complex material
  
  *Susceptibility variations degrade spectral resolution*
“A man with an unusually tiny brain managed to live an entirely normal life despite his condition, caused by a fluid buildup in his skull, French researchers reported … "He was a married father of two children, and worked as a civil servant," Dr. Lionel Feuillet wrote in a letter to the Lancet medical journal.

The man had a shunt inserted into his head to drain hydrocephalus -- water on the brain -- as an infant...the man had an IQ of 75, below average but not considered mentally retarded or disabled, either.”
What is “Molecular Imaging?”

- Uses molecular specific signatures to image function instead of structure
  
  Positron emission tomography (e.g. $^{18}$F, $t_{1/2} = 2$ h); SPECT ($\gamma$ emitters, e.g. $^{99m}$Tc, $t_{1/2} = 6$ h)
  
  Magnetic Resonance Imaging
  
  Optical Imaging (both surface and deep tissue)

- Molecular Medicine: the real opportunity
  
  Cost to bring a drug to market >$1B-almost all in clinical trials

- Society for Molecular Imaging meeting
  
  1st: Boston, 2002 (500 attendees)

- World Molecular Imaging Congress
  
  13th: Seoul, 2014 (3000 attendees)
Three approaches to practical molecular information in optical and MR imaging

1. Study what is already removed:

Thin sample imaging: new molecular contrast in melanoma histology

Cross-phase modulation imaging through fs pulse shaping: nonlinear phase contrast

Bright field

XPM
Three approaches to practical molecular information in optical and MR imaging

- 2. Make better use of intrinsic contrast
  Nonlinear optics: fs pulse shaping/ pulse train modulation
Three approaches to practical molecular information in optical and MR imaging

- 3. Add back what was already there

*Hyperpolarized MR imaging with long-lived states*

Gray: human prostate MR images  
Color: high lactate/pyruvate ratio after injection of HP $^{13}$C pyruvate (UCSF /GE).

Our work: families of MR agents or tags with many-minute lifetimes and biocompatibility
NMR and MRI - the fundamental challenges

- Weak signal: $\approx 10^{-5}$ fractional magnetization
  
  *Detecting metabolites hard (particularly unlabeled)*
  
  *Imaging anything but water difficult*

- Water images structure more than function
  
  *Density, $T_1$, $T_2$, $T_2^*$, diffusion: physics of 1954*

- Tissue - the ultimate complex material
  
  *Susceptibility variations degrade spectral resolution*

- Want to do better? *Use different spin physics*
Hyperpolarization

- Dynamic Nuclear Polarization

- Transfer polarization from electrons to protons at low temperature
- Very general method
Hyperpolarization is one of the most exciting new frontiers in MRI.

Analysis of Cancer Metabolism by Imaging Hyperpolarized Nuclei: Prospects for Translation to Clinical Research

Phase I/II clinical trial going on now with GE/UCSF

New GE clinical polarizer: 10x output, new dose every hour
Clinical applications of hyperpolarization: the potential and challenges

- Delivery in vivo: excellent progress
  
  *GMP system demonstrated*
  
  *Several clinically relevant molecules detected in vivo*

- Creating hyperpolarization: expensive
  
  *Many different molecules hyperpolarized*
  
  *DNP complex and slow (cooling to 1K needed; hour to polarize; >$20M for a clinical system)*

  (Talk this afternoon, S12 (16:50) solves this problem)

- General application: limited by spin physics
  
  *Carbon signal lifetime ca. 1 min in favorable cases*
  
  *Creates great demands on delivery systems*
  
  *Limits accessible biochemical pathways*

This talk: how do you preserve large population?
Pulse sequences in NMR are very powerful, but…

- Removing inhomogeneous broadening ($T_2^*$)
  
  *Spin echo (1950, Hahn); multiple echoes (1955-1956, Carr, Purcell, Meiboom, Gill)*

- Altering effective Hamiltonian ("static" $T_2$)
  
  *Dipolar refocusing (1967, Waugh), time reversal (1971, Pines), two-quantum Hamiltonian (1979, $W^2$)*

- Dozens of QC/QIP papers (slowly varying $T_2$)
  
  *Example: dynamic decoupling (Uhrig 2007) removes effect of low-frequency fluctuations in MRI (2009, $W^2$)*

- None of this work preserves populations ($T_1$)
Two magnetically equivalent spins give a disconnected, long-lived singlet eigenstate.

\[
T_0 \equiv \frac{\alpha\beta + \beta\alpha}{\sqrt{2}}
\]

\[
T_1 \equiv \alpha\alpha
\]

\[
T_{-1} \equiv \beta\beta
\]

\[
S \equiv \frac{\alpha\beta - \beta\alpha}{\sqrt{2}}
\]

Disconnected, "invisible" state

Identical spins (\(\text{CH}_2=\text{CCl}_2\))

Two different spins (\(\text{CHCl}=\text{CHBr}\))
Hyperpolarization and singlet states offer a potential route to long-lived tracers

Singlet state between *inequivalent spins* offered the first approach to long-lived signal (Levitt et. al. *PRL* 2004, 92, 153003; *JACS* 2004, 126, 6228).

Two challenges:

1. *Inequivalent spins require spin locking or zero field*
   
   Sequence in original spin locking paper: 300 MHz head coil, power dissipation (SAR) is 6000 times legal limit.

2. *Very sensitive to paramagnetic relaxation*

   Singlet between *chemically equivalent* spins avoids decoupling; self-sustained at any field strength (Warren et. al. *Science* 2009)

   *Challenge: required chemistry or field cycling to unlock*
The field has converged: symmetry (or near-symmetry), low-\(\gamma\), low field best

- **Levitt:** single pair of near equivalent spins: \(\Delta\omega << 2\pi J_{CC}\)

Magnetization to Singlet to Magnetization (MSM)

- **Warren:** multiple pairs of chemically equivalent spins: \(\Delta\omega = 0\) while \(\Delta J_{CH} \neq 0\).


Can we access the singlet state, without breaking chemical equivalence?

- Chemical equivalence gives a lot of advantages

  *Active decoupling not needed-reduced power dissipation (clinically acceptable)*

  *Relaxation mechanisms have to break symmetry. If environment is the same, they are reduced*

- Doesn’t this violate the laws of physics?

  *No, if the spins are chemically equivalent, but coupled differently to others*
There is nothing new about $A_2B_2$ spin systems—but they are interesting

THE ANALYSIS OF NUCLEAR MAGNETIC RESONANCE SPECTRA

II. TWO PAIRS OF TWO EQUIVALENT NUCLEI

J. A. Pople, W. G. Schneider, and H. J. Bernstein

ABSTRACT

This paper deals with the analysis of the nuclear magnetic resonance spectrum of two pairs of two equivalent nuclei (of spin $\frac{1}{2}$) whose relative chemical shift is of the same order as the spin-coupling constants of the system ($A_2B_2$ in the notation of Part I). The complete matrix is set up and correlated with the results of McConnell, McLean, and Reilly (4) for the corresponding theory with large chemical shifts.

The proton resonance spectrum of naphthalene is reported and is analyzed as an $A_2B_2$ system on the hypothesis that spin couplings between protons on different rings are negligible. A complete analysis of the spectrum of o-dichlorobenzene, which represents a further example of an $A_2B_2$ system, is also given. The spectrum of 1-chloro-2-bromoethane at room temperature is also analyzed as an $A_2B_2$ system.

Example: diacetylene. Today we call this $AA'BB'$ or $AA'XX'$ to accommodate multiple couplings
What are the basis states for AA’BB’?

\[
K = J_A + J_B, \quad L = J - J', \\
M = J_A - J_B, \quad N = J + J'.
\]

<table>
<thead>
<tr>
<th>Function*</th>
<th>( A )</th>
<th>( B )</th>
<th>Diagonal matrix elements†</th>
<th>Off-diagonal matrix elements</th>
</tr>
</thead>
<tbody>
<tr>
<td>( s_1 )</td>
<td>( \alpha \alpha )</td>
<td>( \alpha \alpha )</td>
<td>( \eta H_0(2 - \sigma_A - \sigma_B) + \frac{1}{2}N )</td>
<td>( (1s_1 \left</td>
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<td>( 1s_1 )</td>
<td>( 2^{-\frac{1}{2}}(\alpha\beta + \beta\alpha) )</td>
<td>( \alpha \alpha )</td>
<td>( \eta H_0(1 - \sigma_B) )</td>
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<td>( \alpha \alpha )</td>
<td>( 2^{-\frac{1}{2}}(\alpha\beta + \beta\alpha) )</td>
<td>( \eta H_0(\sigma_A - \sigma_B) - \frac{1}{2}N )</td>
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<td>( 1s_0 )</td>
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<td>( \eta H_0(-\sigma_A + \sigma_B) - \frac{1}{2}N )</td>
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<td>( 2s_0 )</td>
<td>( \alpha \alpha )</td>
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<td>( -K )</td>
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<td>( 3s_0 )</td>
<td>( 2^{-\frac{1}{2}}(\alpha\beta - \beta\alpha) )</td>
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<td>( 4s_0 )</td>
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<td>( \eta H_0(-1 + \sigma_B) )</td>
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<td>( \eta H_0(-1 + \sigma_A) )</td>
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<td>( 2^{-\frac{1}{2}}(\alpha\beta + \beta\alpha) )</td>
<td>( \eta H_0(-2 + \sigma_A + \sigma_B) + \frac{1}{2}N )</td>
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<tr>
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<td>( \beta \beta )</td>
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</table>

*Function* represents the electronic configuration of the system.

†Diagonal matrix elements are given by \( \eta H_0 \) with appropriate combination of quantum numbers.

The off-diagonal matrix elements are calculated based on the specific combination of \( A \) and \( B \) functions and their respective quantum numbers, taking into account the selection rules for the system.
Pairs of equivalent spins and couplings create a versatile class of long-lived reagents

- Two carbons and two hydrogens have highly protected singlet-singlet state \((\alpha\beta-\beta\alpha)_C(\alpha\beta-\beta\alpha)_H\)

- Triplet-triplet state \((\alpha\beta+\beta\alpha)_C(\alpha\beta+\beta\alpha)_H\), accessible from thermal magnetization or hyperpolarization, has the same symmetry.

- If \(J_{CC}+J_{HH}>J_{CH}-J'_{CH}\) SS and TT stable:

\[
H / 2\pi\hbar = \begin{pmatrix}
TT & \left( \frac{1}{2}(J_{CC}+J_{HH}) , - \frac{1}{2}(J_{CH}-J'_{CH}) \right) \\
SS & \left( - \frac{1}{2}(J_{CH}-J'_{CH}) , - \frac{1}{2}(J_{CC}+J_{HH}) \right)
\end{pmatrix}
\]

- To invert spins with a weak pulse, go on resonance. Here that means give many 180s to only C or only H at the right rate:

\[
\tau = 1 / \left( 2\sqrt{(J_{CC}+J_{HH})^2 + (J_{CH}-J'_{CH})^2} \right)
\]

\[
n = \pi / \left( 2\arctan((J_{CH}-J'_{CH}) / (J_{CC}+J_{HH})) \right)
\]
Inversion symmetry seems to be valuable experimentally

- Azobenzene: trans $\rightarrow$ cis with light
  \[ T_1(\text{nitrogen}) = 8 \text{ sec (8.4 T)} \]
  \[ \text{Trans } T_s: 60 \pm 10 \text{ s; cis } T_s: < T_1 \]

- Diphenylacetylene (8.4T):
  \[ T_s > 300 \text{ s; insensitive to oxygen} \]
Long-lived “markers” as biochemical probes

**Current Work**

- Attaching diphenyl-$^{13}$C$_2$-acetylene e.g. onto suberoylanilide hydroximimic acid (SAHA), a histone deacetylase inhibitor and anti-cancer drug.

- Classes of drugs that inherently contain diphenyl-$^{13}$C$_2$-acetylene segments. For example, antibiotic LpxC inhibitors based on a diphenyl-diacetylene (1,4-diphenyl-1,3-butadiyne)-threonylhydroxamate scaffold.

Singlet polarization lifetime measurements at 8T of $^{13}$C$_2$-MDPA (light-blue) and $^{13}$C$_2$-DPA (dark-blue). Longitudinal magnetization relaxes with $T_1$=12 s for both compounds, as indicated by the green line.
Generality: biologically relevant molecules and molecular tags

- Phenbutazone (animal NSAID)
- L-Dopa
- Naphtalenes, naphthoquinonones, pyridazines
- Maybe separated $^{14}$N (pyrazines, adenine?)

C. J. Lee et al., Chem Biol 18 (2011).
E. Christiansen et al., J Med Chemistry 56 (2013).
How general is this approach?

**AA’X₂X₂’: many different two-level systems**

<table>
<thead>
<tr>
<th>Γ'</th>
<th>∑mₜ</th>
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<tr>
<td>+1</td>
<td></td>
<td>( \frac{1}{\sqrt{2}} (T_{+1}T_0 + T_0T_{+1})T_0 )</td>
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<tr>
<td>0</td>
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<td>( \frac{1}{\sqrt{2}} (T_{-1}T_{+1} + T_{+1}T_{-1})T_0 )</td>
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<td>+1</td>
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<tr>
<td>-1</td>
<td></td>
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<td></td>
<td>( \frac{1}{\sqrt{2}} (T_{-1}S - ST_{-1})T_0 )</td>
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</tbody>
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Eight have the same resonance condition; the two “obvious” states (in red) are different.
How general is this approach?

- AA’QQ” where X is spin 1 (D, $^{14}$N)

\[ J_n = 54 \text{ Hz} \quad J_f = -6.3 \text{ Hz} \]
\[ J_{CC} = 154 \text{ Hz} \]
\[ T_S = 49 \pm 23 \text{s} \]

\[ J_n = 8.2 \text{ Hz} \quad J_f = -0.9 \text{ Hz} \]
\[ J_{CC} = 154 \text{ Hz} \]
\[ T_S = 83 \pm 30 \text{ s} \]

$^{13}$C $T_1 = 16.7 \pm 0.9$ s, $^2$H $T_1 = 0.5 \pm 0.2$ s

Deuterated DPA: 500 s at 9.4T (protonated 310 s). All are much longer at 1T.
Opinion: HP MRI will have broadest impact at low fields with multimodality imaging

- Conventional NMR: $\text{SNR} \propto \gamma_{\text{prep}} \gamma_{\text{det}}^{7/4} B^{7/4}$
  
  Boltzmann equilibrium $\propto \gamma_{\text{prep}} B$; Magnetization $\propto \gamma_{\text{det}}$;
  Induced voltage $\propto \omega = \gamma_{\text{det}} B$; coil noise $\propto \omega^{1/4}$

- Conventional MRI: $\text{SNR} \propto \gamma_{\text{prep}} \gamma_{\text{det}} B^1$
  
  Coil noise $\propto \omega$, dominated by body

- Hyperpolarized MRI: $\text{SNR} \propto \gamma_{\text{det}}$
  
  Boltzmann factor eliminated!

- Relaxation times usually much longer at low field; power dissipation lower ($\text{SAR} \propto B^2$)
  
  Compromise (0.5-1.5T) to get a good anatomic and HP image, or marry with CT for even lower field
Relevant papers and Extraordinary People


Other links: www.cmbi.duke.edu

MR: Dr. Thomas Theis, Yesu Feng, Kevin Claytor, Ryan Davis, Zijian Zhou, Jin Yu

NIH, NSF