Sampling

David Rovnyak
4’th Biomolecular SSNMR Winter School
Stowe, VT, 2016
## Acknowledgements

<table>
<thead>
<tr>
<th>Location</th>
<th>Acknowledgments</th>
</tr>
</thead>
<tbody>
<tr>
<td>Bucknell</td>
<td>Levi Craft, Melissa Palmer, Riju Gupta, Mark Sarcone, Ze Jiang</td>
</tr>
<tr>
<td>Delaware</td>
<td>Tatyana Polenova, Chris Suiter</td>
</tr>
<tr>
<td>UCHC</td>
<td>Jeffrey C. Hoch</td>
</tr>
<tr>
<td>UVA</td>
<td>James Rovnyak, Virginia Rovnyak</td>
</tr>
<tr>
<td>Susquehanna</td>
<td>Geneive Henry</td>
</tr>
<tr>
<td>Funding</td>
<td>NSF-RUI, NIH-AREA</td>
</tr>
<tr>
<td>Thank you’s</td>
<td>Adam Schuyler (UCHC), Frank Delaglio (NIST/UMD)</td>
</tr>
</tbody>
</table>

Big thank you to Mei Hong and Chris Jaroniec and Sponsors.
Thank You
Goals/Outcomes

I. Interesting foundations of sampling/ Classic Fourier Calcs

II. NUS fundamentals; “good” NUS schedules

III. How to calculate a NUS sensitivity enhancement

IV. Speculations on frontiers in sampling
I. Interesting Foundations

Aiming for key aspects of sampling which are assumed/skimmed in other texts.

- The Fourier Transform and Fourier Pairs
- Parseval’s Theorem: the FT is power conserving
- Noise in the FID and the frequency spectrum
- Signal in the FID and the frequency spectrum
- Signal to Noise Ratio in the time domain
Signal-to-Noise and Sensitivity

**SNR**: ratio of the peak maximum to the rms noise.

\[ \text{rms} = \left[ \frac{1}{N} \sum_{i=1}^{N} s(\omega_i)^2 \right]^{\frac{1}{2}} \]

**Sensitivity**: ratio of the SNR to the square root of total time (Ernst)

\[ \frac{S(\omega_0)}{\text{rms noise}} \]

\[ \frac{\text{SNR}}{\sqrt{\text{time}}} \]

**P-Set 1.6**
Noise is white gaussian

- Thermal noise in a copper wire theoretically “white gaussian”
  - white: all frequencies occur; no holes
  - gaussian: intensity of a noise peak follows a Gaussian distribution

![Diagram with signal and simulated white gaussian noise comparison](image)

- Spectrometer noise
- Simulated white gaussian noise

*is this signal or a Gaussian excursion?*
Noise and temperature

Noise measured with temperature of a 50 Ohm load (Glenn Facey)
Complex Fourier Transform

\[ S(\omega) = \int_{-\infty}^{\infty} S(t) e^{-i2\pi \omega t} \] \hspace{1cm} (1)

\[ S(t) = \int_{-\infty}^{\infty} S(\omega) e^{i2\pi \omega t} \] \hspace{1cm} (2)
The FT is Linear

\[ S(\omega) = \int_{-\infty}^{\infty} (f(t) + g(t))e^{-i2\pi\omega t} = \int_{-\infty}^{\infty} f(t)e^{-i2\pi\omega t} + \int_{-\infty}^{\infty} g(t)e^{-i2\pi\omega t} = f(\omega) + g(\omega) \]
Discrete Fourier Transform

\[ f \left( \omega_k \right) = \sum_{j=0}^{N-1} s_j(t) e^{-\frac{i}{N} jk} \]

Precession of \( M \) induces current in coil

Fourier transform

Signal digitized and stored in a computer

\( V_{\text{coil}} \)

\( s(t) \)

\( \nu \)

\( L \)

frequency

time
Nyquist frequency, \( v_{\text{max}} \): let \( v_{\text{max}} \) be the highest frequency you wish to detect in units of Hz (s\(^{-1}\)). Then you must digitize 2\( v_{\text{max}} \) times per second. This requires collecting two points per wavelength \( \lambda_{\text{max}} \).

**dwell time**, \( \Delta t \): time interval between points; \( \Delta t = 1/(2v_{\text{max}}) \); \( v_{\text{max}} = 1/(2\Delta t) \)

no sign discrim.

\[
\text{sw} = v_{\text{max}}
\]

with sign discrim.

\[
\text{sw} = 2v_{\text{max}}
\]

NMR spectral width, part 2: sign discrimination detects frequencies from - \( v_{\text{max}} \) to + \( v_{\text{max}} \) and so the NMR spectral width is \( 2v_{\text{max}} = 1/(\Delta t) \).

Example: \( \Delta t = 20 \mu s \), then \( \text{sw}_{\text{NMR}} = 50 \text{ kHz} \)
The Discrete Fourier Transform is Cyclic …

F. Delaglio
The Discrete Fourier Transform is Cyclic …
As Signals Get Faster Than The Sampling Frequency …
Folding

Folding is circular.

How far out can you fold? Filters remove noise (and signal) typically above 1.1-1.3*$v_{max}$.

Coming up: NUS and spectral width.
DFT and Resolution

acquisition time, $t_a$: this is $N\Delta t$, where $N$ is the number of complex points

digital resolution, $R_d$: the frequency separation between points in the spectrum $R_d = 1/t_a$

$R_d >> \text{LW}$  $R_d \sim \text{LW}$  $R_d << \text{LW}$

Zerofilling improves $R_d$ without distorting the spectrum:

Why is this legal? Roughly: appending 0’s is adding no data. Formally: Parseval.
Let’s see the DFT in action.

- Credits and thanks: Frank Delaglio
\[ x(f) = \sum x(t) \left[ \cos \left( \frac{2\pi f t}{N} \right) - i \sin \left( \frac{2\pi f t}{N} \right) \right] \]

Each Fourier term corresponds to a point in the spectrum.
First, Let’s View the Fourier Transform with the Imaginary Parts Hidden …
Time Domain (Real Part)

Fourier Term

Multiplied with Time Domain Data

Forms the Product
When the Fourier term does not match any frequency in the data, the product has balanced amounts of positive and negative intensity, and sums to zero.
Time Domain (Real Part)

Fourier Term

Multiplied with Time Domain Data

Forms the Product

\[ S^+ \quad S^- \]

Sum Over Product to Form a Frequency Point:

Frequency Domain (Real Part)
Now, Let’s View the Fourier Transform with the Both the Real and Imaginary Parts Shown …
**Time Domain**

Complex Fourier Term

Multiplied with Complex Time Domain Data

Forms the Complex Product

Sum Over Product to Form a Frequency Point:

**Frequency Domain**
Time Domain

Complex Fourier Term

Multiplied with Real-Only Time Domain Data

Forms the Complex Product

Sum Over Product to Form a Frequency Point:

Frequency Domain
**Time Domain**

Complex Fourier Term

Multiplied with Imaginary-Only Time Domain Data

Forms the Complex Product

Sum Over Product to Form a Frequency Point:

**Frequency Domain**
Combined, the Real and Imaginary Parts Distinguish Between Positive and Negative Frequencies …
In Biomolecular NMR, Indirect Dimensions are Often Truncated and Have Limited Numbers of Points ...
Since the Fourier product is truncated, there is no longer ideal cancelation of positive and negative intensities, giving broad lines and artifacts.
The FT of the product of two functions is related to the FT’s of the individual functions by convolution: the overlap of scanning one function across the other.

\[ F(f 	imes g) = f(\omega) \otimes g(\omega) = F(f) \otimes F(g) \]

(note: horrible typo in original notes)
FT : Theorems

- Convolution
- Parseval
- Nyquist

\[ F(f \ast g) = f(\omega) \otimes g(\omega) \]

\[
\begin{align*}
 f(t) \times g(t) &= f(t) \quad \text{and} \quad g(t) \\
 \text{FT} &= f(\omega) \otimes g(\omega)
\end{align*}
\]
An exponential decay transforms to a Lorentzian

\[
S(\omega) = \int_{-\infty}^{\infty} e^{-t/T_2} e^{-i2\pi\nu t} \, dt = \int_{0}^{\infty} e^{-t(1/T_2 + i2\pi\nu)} \, dt
\]

\[
= \left. \frac{-1}{(1/T_2 + i2\pi\nu)} \right|^{\infty}_{0} e^{-t(1/T_2 + i2\pi\nu)}
\]

\[
= \frac{-1}{(1/T_2 + i2\pi\nu)} \left[ 0 - 1 \right]
\]

\[
= \frac{1}{(1/T_2 + i2\pi\nu)} \frac{1}{(1/T_2 - i2\pi\nu)}
\]

\[
= \frac{1/T_2}{\left(1/T_2^2 + 4\pi^2\nu^2\right)} + \frac{-i2\pi\nu}{\left(1/T_2^2 + 4\pi^2\nu^2\right)}
\]

\[
= \frac{1}{2\pi} \frac{1/2 \, LW}{\left(LW^2 + \nu^2\right)} + \frac{-i2\pi\nu}{\left(1/T_2^2 + 4\pi^2\nu^2\right)}
\]

\[
\text{Real Part} \quad \text{Imaginary Part}
\]

\[
T_2 = 0.010 \text{ s}
\]

\[
LW = \text{FWHH} = \frac{1}{\pi T_2} \text{ (Hz)}
\]
Other Fourier Pairs

- A Gaussian transforms to a Gaussian (not the same one)

\[ S(\omega) = \int_{-\infty}^{\infty} e^{-t^2/2\sigma^2} e^{-i2\pi \nu t} dt = \int_{0}^{\infty} e^{-(t^2/2\sigma^2 + i\omega t)} dt \]

From tables
\[ \int_{-\infty}^{\infty} e^{-ax^2 + bx} dx = e^{\frac{b^2}{4a}} \sqrt{\frac{\pi}{a}} \]

\[ S(\omega) = \frac{1}{\alpha \sqrt{2\pi}} e^{\frac{-\nu^2}{2\alpha^2}}, \quad \alpha = \frac{1}{2\pi \sigma} \]

There are a number of ways to tackle this integral. A fun alternative is to notice that you can turn this into a more conventional Gaussian integral. Consider the exponent:

\[ -t^2/2\sigma^2 - i\omega t \]

Complete the square with some \( b \)

\[ -t^2/2\sigma^2 - i\omega t - b^2 + b^2 \]

Figure out \( b \) to lead you to

\[ \frac{-1}{2\sigma^2} (t + i\omega \sigma^2)^2 - \frac{\omega^2 \sigma^2}{2} \]
Parseval: the FT is Power Conserving

\[
\int_{-\infty}^{\infty} |s(t)|^2 = \frac{1}{2\pi} \int_{-\infty}^{\infty} |s(\omega)|^2
\]

Practical consequence: the FT is like life, what you get out of it, is exactly what you put in to it.
Noise

\[ \text{noise} \propto \sqrt{\text{time}} \]

Analogy to signal averaging.

See for example:
Noise

$^1$H FID

RMS Noise (arbitrary units)

Evolution time (sec)

$f(t) = \sqrt{t}$

Experimental RMS Noise

JBNMR 2004, 30, 1-10
Signal

\[ S(\omega = \omega_0) = \int_0^t e^{i\omega_0 t} e^{-R_2 t} e^{-i\omega t} dt \]

\[ = \int_0^t e^{-R_2 t} dt \]

\[ = T_2 \left(1 - e^{-t_{acq}/T_2}\right) \]
\[ S_N(t) = \frac{T_2 \left(1 - e^{-t/T_2}\right)}{C_N \sqrt{t}} = \frac{\text{peak height}}{\text{r.m.s. noise}} \]
SNR in the Time Domain


The graph of Eq. [9], given in Fig. 1, shows a relatively broad maximum about \( T = 1.26 T_2 \); over 99% of the maximum value is obtained between \( T_2 \leq T \leq 1.5 T_2 \). It is worth noting that this result is independent of the value of the \( S/N \) ratio.

**Fig. 1.** Normalized graph of Eq. [9] as a function of \( T/T_2 \). The function \( f(x) \) is proportional to \( A(T)/\delta Q(T) = (2M_0/PT^{1/2}) [1-exp (-T/T_2)] \), and \( x \) is \( T/T_2 \), where \( T \) is the integration length and \( T_2 \) is the exponential relaxation time constant. The graph shows a broad maximum around \( T = 1.26 T_2 \).
Regarding $1.26 \ T_2$


$$S/N = \frac{M_0 T_2^*}{\sqrt{T_1/2} (1 - e^{-T/T_2^*})}$$


- **T. Vosegaard, N. Chr. Nielsen, 2009.** Defining the sampling space in multidimensional NMR experiments: What should the maximum sampling time be? J. Magn. Reson. 199, 147-158.


Signal Envelope Determines Sensitivity

Sensitivity of Two-Dimensional Spectra

MALCOLM H. LEVITT, GEOFFREY BODENHAUSEN, AND R. R. ERNST

Laboratorium für Physikalische Chemie, Eidgenössische Technische Hochschule,
8092 Zurich, Switzerland

Received January 10, 1984

Fig. 1. Typical signal envelope function $s^e(t_1, t_2)$ which decays exponentially in the two time domains $t_1$ and $t_2$, as described by Eq. [5]. The sensitivity of the 2D experiment depends primarily on the average height of the signal envelope function over the “captive volume” defined by the boundaries $0 < t_1 < t_1^{\text{max}}, 0 < t_2 < t_2^{\text{max}}$.

1984 J Magn Reson 58:462–472
Conventional and Exponential Sampling for 2D NMR Experiments with Application to a 2D NMR Spectrum of a Protein

JENNIFER C. J. BARNA AND ERNEST D. LAUE

Department of Biochemistry, Tennis Court Road, Cambridge CB2 1QW, England

Received July 2, 1987

NMR experiments by considering alternative methods of data acquisition. We have proposed (5, 6) a novel form of selective data sampling which we call exponential sampling. In this method many $t_1$ points would be acquired near the beginning of the experiment where the signal-to-noise ratio is high but some points, acquired with an exponentially decreasing frequency, would be acquired later in the experiment to aid the determination of high-resolution information. Such sampling would be used in


Definitions

Some misconceptions have arisen in the literature – we’ll talk about more of those throughout, but a great deal can be clarified by these definitions, which are suggested for broader adoption.

**Intrinsic SNR (iSNR)**

- SNR of raw data, prior to any manipulation
- Once receiver is off, the iSNR cannot be changed
- May always be computed in time domain
- Equivalent to frequency spectrum for power conserving transform.

**Apparent SNR (SNR)**

- SNR after signal manipulation:
  - post-apodization
  - post-linear prediction
- May be computed in time domain or frequency domain
II. NUS Fundamentals

- On grid NUS
- Weighted NUS
- Point Spread Function and Convolution
- Partial Component; Nonuniform Weighting
- Spectral window of NUS

What is a “good” NUS schedule?

What is good spectral reconstruction?
Preaching to converted, but…why NUS?

Comment: building trust with non-Fourier spectral estimation

Most common approach
Samples are a subset of the evenly (uniformly) spaced Nyquist grid.
Equal number of transients for each sample.
Weighted Sampling

NUS and NUWS have same density of samples.

Weighted Sampling (NUS)

Weighted averaging (NUWS)

Sampling Density

(Kumar et al., *JMR 95*, 1-9, 1991)
Aside: Processing Weighted Averaging

NUWS (weighted averaging) is on-grid so the FFT may be used.

But NUWS introduces the apparent line broadening of the sampling density.

The FFT will be power conserving, fast, and easy, but introduces this broadening.

Non-Fourier techniques avoid the broadening of the sampling density. (just like NUS)

Perhaps applicable in imaging where lines are very broad.

Some other NUWS works.


Exponential Weighted Sampling

![Graph showing exponential weighted sampling with signal intensity on the y-axis and evolution time on the x-axis. The graph includes a line and data points indicating exponential decay.]
Aside: Random Sampling, Random Order

<table>
<thead>
<tr>
<th>Conventional Uniformly Sampled (US) Schedule</th>
<th>Non-Uniform Sampling Schedule</th>
<th>Non-Uniform Sampling Schedule in Random Order</th>
</tr>
</thead>
<tbody>
<tr>
<td>0   1   2   3   4   5   6   7   8   9   10  11  12  13  14  15</td>
<td>0   1   3   5   8   10   11   5   8   3   15   10</td>
<td>0   1   11   5   8   3   15   10</td>
</tr>
</tbody>
</table>

Random order:
- A high res spectrum develops in real time.
- Allows for time resolved NMR, for example:

```
10 5 9 23 3 31 7 17 11 2 28 16 4 32 24 1 19 12 28 8 21 3 32 2 16 .....```
Aside: Random Sampling, Random Order

(Mayzel, Rosenlow, Isaksson, Orekhov, JBNMR 2014, 58: 129-139)
Convolution and Point Spread Function

PSF – the Fourier transform of the sampling schedule

Point Spread Function

What is a good PSF?

Larger gaps

Smaller gaps between samples

Low noise around central feature corresponds to more robust reconstructions

(aside – ignore stray horizontal line)
Partial Component NUS

Example NUS sampling schedule for 3D NMR; NUS in both $t_1$ and $t_2$.

$$(t_1, t_2) = \text{RR, RI, IR, II}$$

Proposal: select RR,RI,IR,II spectra nonuniformly as well.
Partial Component NUS

Summary: PC is a route to introducing randomness, may help improve PSF’s with sparsity.
## Spectral Window of NUS

**On grid NUS breaks Nyquist.**

**Main concern is to avoid aliasing within the spectral window.**

<table>
<thead>
<tr>
<th></th>
<th></th>
</tr>
</thead>
<tbody>
<tr>
<td>In noiseless spectra, the GCD translates in principle in to the exact spectral window.</td>
<td></td>
</tr>
</tbody>
</table>

| On Grid: as long as sampling not too sparse, the effective spectral window corresponds closely to the Nyquist frequency of the original grid; |                                                                                                  |
| Proposal – choose NUS from oversampled Nyquist grid guideline - BURSTY samples (some uniform tracts) |                                                                                                  |
Example of selecting NUS from an oversampled grid.

Don’t run for the hills – notice the cross-sections – small effect but worth knowing.
**Good schedule? Processor?**

<table>
<thead>
<tr>
<th>Random</th>
<th>PSF’s, Gapping, Partial Component</th>
</tr>
</thead>
<tbody>
<tr>
<td>Coverage</td>
<td>Non-sparse: 25-50% reduction</td>
</tr>
<tr>
<td></td>
<td>Sparse: &lt; 25% reduction</td>
</tr>
<tr>
<td>Weighted</td>
<td>Sensitivity, Resolution</td>
</tr>
<tr>
<td>Reproducibility</td>
<td>Reporting, validating, regulatory issues</td>
</tr>
<tr>
<td>Gridding for SW</td>
<td>Minimizing GCD of samples</td>
</tr>
</tbody>
</table>

In order to focus on fundamentals of sampling, processing is not the focus here. To make a long story short:

Processing: CS, L1, L2, hmsIST, IST, MaxEnt (MINT, FM, more), MDD, more.

While improvements remain, reasonably modern implementations of ALL of these are normally robust for non-sparse NUS. In contrast, best processing for sparse NUS remains a going concern.
Calculate a NUS Enhancement

- Working through an example
- Exact Theory for several cases
- Two Theorems for NUS

Fundamentals: Parseval
Example: NUS Enhancement

- Let’s see a practical example before looking at the theory.
An FID

Evolution time / $T_2$
Uniform Sampling

32 uniform samples

Evolution time / $T_2$
Sum = 9.97
NUS 8/32 2X bias exponential

NUS 8/32 1.5X bias exponential

NUS 8/32 matched exponential

Uniform 32 samples
Suppose the uniform experiment used 4 transients for each of 32 increments, for

\[ 4 \times 32 = 128 \text{ transients} \quad \text{(uniform)} \]

Then NUS must use **fourfold** more, or 16 transients for each of 8 increments, for

\[ 16 \times 8 = 128 \text{ transients} \quad \text{(NUS)} \]
NUS 8/32 2X bias exponential

NUS 8/32 1.5X bias exponential

NUS 8/32 matched exponential

Uniform 32 samples
Matched NUS

Sum = 17.79
Recall that the uniform experiment and the NUS experiment each used 128 transients.

Noise is the same in each experiment.
Enhancement: Match NUS

- Enhancement for acquisition spanning $3.14 T_2$ and with NUS probability density matched to $T_2$:

$$\begin{align*}
\text{Signal Sum by NUS} & = 17.79 \\
\text{Signal Sum by Uniform} & = 9.97 \\
\text{Computed by exact theory:}^* & = 1.78
\end{align*}$$

- Computed by exact theory:*  

\[ \text{enhance} = 1.71 \]

*MRC, V49, 483-491, 2011
Uniform 32 samples

NUS 8/32 2X bias exponential

NUS 8/32 1.5X bias exponential

NUS 8/32 matched exponential
1.5X Bias NUS

Sum = 19.57
Enhance = 19.57/9.97 = 1.96
Theory = 2.00
Slight discrepancy?

<table>
<thead>
<tr>
<th></th>
<th>match</th>
<th>match</th>
<th>match</th>
</tr>
</thead>
<tbody>
<tr>
<td>compute</td>
<td>1.78</td>
<td>1.96</td>
<td>2.07</td>
</tr>
<tr>
<td>theory</td>
<td>1.71</td>
<td>2.00</td>
<td>2.20</td>
</tr>
</tbody>
</table>
Exact Theory – How does this work?

Double Dipping: Enhance SNR by NUS

1. Eliminate samples $> 1.26T_2$
   Decrease Noise.

2. Use time savings to increase transients for all remaining samples.
   Increase Signal.

(Double Dipping: Enhance SNR by NUS)
Optimization of Two-Dimensional NMR by Matched Accumulation

ANIL KUMAR,* †‡ STEPHEN C. BROWN,* MARY E. DONLAN,* BEAT U. MEIER,§ AND PETER W. JEFFS*

*Department of Structural and Biophysical Chemistry, Glaxo Research Laboratories, 5 Moore Drive, Research Triangle Park, North Carolina 27709; †Department of Chemistry, University of North Carolina, Chapel Hill, North Carolina 27514; and §Bruker Instruments Inc., Manning Park, Billerica, Massachusetts 01821

Received August 21, 1990; revised April 22, 1991

form dependent on the coherence detected. Matching the time spent signal averaging to the expected amplitude of the signal observed should also improve the detected signal-to-noise. Following this reasoning, Barna et al. (J. Magn. Reson. 75, 384, 1987) demonstrated the utility of exponential sampling in one- and two-dimensional NMR, using maximum-entropy methods to analyze the data. It is proposed here that for two-dimensional experiments the exponential sampling be replaced by exponential averaging. The data thus

<table>
<thead>
<tr>
<th>Time</th>
<th>Area (exp.)/area (const.)</th>
<th>Gain in S/N</th>
</tr>
</thead>
<tbody>
<tr>
<td>$1 \times T_2$</td>
<td>0.63</td>
<td>1.26</td>
</tr>
<tr>
<td>$2 \times T_2$</td>
<td>0.43</td>
<td>1.50</td>
</tr>
<tr>
<td>$3 \times T_2$</td>
<td>0.32</td>
<td>1.77</td>
</tr>
</tbody>
</table>

(Kumar et al., JMR 95, 1-9, 1991)
Step 1: Enforce Equal Experiment Times

Sample Density (a.u.)

Force area of $h(t)$ equal to area of uniform with a scaling factor $\chi$.

$$\chi(t_{\text{max}}) = \frac{t_{\text{max}}}{\int_0^{t_{\text{max}}} h(t) \, dt}$$

$$\chi_{\text{match-exp}} = \frac{\alpha_{\text{max}}}{1 - e^{-\alpha_{\text{max}}}}$$
Step 2: Form Ratio Of US to NUS Signal

For any nonuniform sampling density $h(t)$ applied to an exponentially decaying signal:

$$
\eta = \frac{\int_0^{t_{\text{max}}} \chi h(t) e^{-t/T_2} dt}{\int_0^{t_{\text{max}}} e^{-t/T_2} dt} = \frac{\chi \int_0^{t_{\text{max}}} h(t) e^{-t/T_2} dt}{T_2 \left(1 - e^{-t_{\text{max}}/T_2}\right)}
$$
Cartoon Proof of Enhancement

\[ s(t) \times e^{-t/T_2} = \text{Signal}_{\text{Uniform}} \]

\[ s(t) \times e^{-t/T_{\text{SMP}}} = \text{Signal}_{\text{NUS}} \]
## Example Enhancements

<table>
<thead>
<tr>
<th>$t_{max}/t_2$</th>
<th>(T_2/T_{smp})</th>
<th>0.5</th>
<th>1.0</th>
<th>1.5</th>
<th>2.0</th>
<th>2.5</th>
</tr>
</thead>
<tbody>
<tr>
<td>0.5</td>
<td></td>
<td>1.01</td>
<td>1.02</td>
<td>1.03</td>
<td>1.04</td>
<td>1.05</td>
</tr>
<tr>
<td>1.0</td>
<td></td>
<td>1.04</td>
<td>1.08</td>
<td>1.12</td>
<td>1.16</td>
<td>1.20</td>
</tr>
<tr>
<td>1.5</td>
<td></td>
<td>1.09</td>
<td>1.18</td>
<td>1.27</td>
<td>1.34</td>
<td>1.41</td>
</tr>
<tr>
<td>2.0</td>
<td></td>
<td>1.16</td>
<td>1.31</td>
<td>1.45</td>
<td>1.57</td>
<td>1.67</td>
</tr>
<tr>
<td>2.5</td>
<td></td>
<td>1.24</td>
<td>1.48</td>
<td>1.67</td>
<td>1.83</td>
<td>1.95</td>
</tr>
<tr>
<td>3.0</td>
<td></td>
<td>1.34</td>
<td>1.66</td>
<td>1.92</td>
<td>2.12</td>
<td>2.26</td>
</tr>
<tr>
<td>3.14</td>
<td></td>
<td>1.37</td>
<td>1.71</td>
<td>2.00*</td>
<td>2.20</td>
<td>2.35</td>
</tr>
<tr>
<td>3.5</td>
<td></td>
<td>1.45</td>
<td>1.86</td>
<td>2.18</td>
<td>2.41</td>
<td>2.58</td>
</tr>
</tbody>
</table>
Exponential NUS-based enhancement

<table>
<thead>
<tr>
<th>Spectrum</th>
<th>Residue</th>
<th>Correlation</th>
<th>Intensity Enhancement*</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td></td>
<td>N-Cα</td>
<td>NUS1 (NUS2)</td>
</tr>
<tr>
<td>2D NCA</td>
<td>Met</td>
<td>N-Cα</td>
<td>2.14 (1.82)</td>
</tr>
<tr>
<td></td>
<td>Leu</td>
<td>N-Cα</td>
<td>2.06 (1.82)</td>
</tr>
<tr>
<td></td>
<td>Phe</td>
<td>N-Cα</td>
<td>2.01 (1.81)</td>
</tr>
<tr>
<td>2D NCACX</td>
<td>Met</td>
<td>N-Cα</td>
<td>2.11 (1.71)</td>
</tr>
<tr>
<td></td>
<td></td>
<td>N-Cβ</td>
<td>2.16 (1.58)</td>
</tr>
<tr>
<td></td>
<td></td>
<td>N-Cβ</td>
<td>1.93 (1.52)</td>
</tr>
<tr>
<td></td>
<td></td>
<td>N-Cβ</td>
<td>2.23 (1.74)</td>
</tr>
<tr>
<td></td>
<td></td>
<td>N-Cβ</td>
<td>1.97 (1.77)</td>
</tr>
<tr>
<td></td>
<td></td>
<td>N-Cβ</td>
<td>2.02 (1.72)</td>
</tr>
<tr>
<td></td>
<td></td>
<td>N-Cβ</td>
<td>1.98 (1.63)</td>
</tr>
<tr>
<td></td>
<td></td>
<td>N-Cβ</td>
<td>1.92 (1.54)</td>
</tr>
<tr>
<td></td>
<td></td>
<td>N-Cβ</td>
<td>1.95 (1.60)</td>
</tr>
<tr>
<td>Phe</td>
<td>N-Cα</td>
<td></td>
<td>1.93 (1.62)</td>
</tr>
<tr>
<td>Phe</td>
<td>N-Cβ</td>
<td></td>
<td>2.03 (1.80)</td>
</tr>
<tr>
<td>Phe</td>
<td>N-CO</td>
<td></td>
<td>2.06 (1.53)</td>
</tr>
</tbody>
</table>

Verified with MINT*

* Power Conserving Regime of MaxEnt
* Non sparse
* large lambda

(Paramasivam, Suiter, Sun, Palmer, Hoch, Rovnyak, Polenova, JPC B, V116, 7416-7427, 2012.)
Gaussian NUS

Some Gaussian NUS:


Trade-Off?

Sample Number

Evolution time / $T_2$

Signal (a.u.)

Exponential NUS BIAS

Uniform, FFT

Sample Number

$15N$ (ppm)

Trade-Off?

(\textit{JBNMR} 58, 303, 2014)
NUS Density and Line Shapes

Matched Exp: enhance = 1.7

Sinusoid: enhance = 1.7

More samples by sinusoid density

Sampling Density

time/T₂

(JBNMR 58, 303, 2014)
Improved Peak Base: Sine vs. Matched Exp

Uniform

Sine

Match Exp.

(JBNMR 58, 303, 2014)
Compound SNR

3D – biosolids NMR: measured enhancements 2.7 to 3.3

(Paramasivam, Suiter, Sun, Palmer, Hoch, Rovnyak, Polenova, JPC B, V116, 7416-7427, 2012.)
BioSolids NMR

Key: already operating at long evolution times in multiple indirect dimensions.

Best positioned to take advantage of compounded NUS enhancements since CT periods less common.

Towards stronger statements about NUS sensitivity

(Palmer et al, J. Phys. Chem. 2015)
FFT of Simulated NUWS data

(a) NUS: match

(b) NUS: 2x bias

(_fft of simulated NUWS data (Palmer et al., J. Phys. Chem. 2015))
EXPERIMENTS

Very little improvement due to NUS at 1.26 $T_2$

(Palmer et al, J. Phys. Chem. 2015)
Experiments

Improvement due to NUS after 1.26 $T_2$

(Palmer et al, J. Phys. Chem. 2015)
Validation with MINT

Is the enhancement always greater than 1?
Theorem I (NUS Sensitivity Theorem). Assume $h(t)$ is a positive, nonincreasing function on some interval $0 \leq t < A$, where $A \leq \infty$. For any positive $T_2$ and $0 < t_{\text{max}} < A$, the time domain iSNR enhancement satisfies

$$
\eta = \frac{\alpha}{\left(1 - e^{-\alpha}\right)} \frac{\int_0^{t_{\text{max}}} h(t)e^{-t/T_2} \, dt}{\int_0^{t_{\text{max}}} h(t) \, dt} \geq 1 ,
$$

where $\alpha = t_{\text{max}}/T_2$, and equality holds if and only if $h(t)$ is constant for $0 < t < t_{\text{max}}$.

Proving the equality statement settles something important: unweighted NUS (sometimes called random NUS) has equal iSNR to uniform sampling.
Theorem 2 (Matched NUS SNR Theorem): The iSNR of matched exponential NUS always has a positive slope for an arbitrary, positive $T_2$ and positive evolution time $t_{\text{max}}$.

Narrower scope, but means:

Exponential NUS simultaneously improves resolution and iSNR with additional experimental time.
Examples of Theorem 1

normalized probability densities

SINE

GAUSS

SNR enhancement by NUS
Examples of Theorem 1

(normalized probability densities)

LINEAR

COSINE

SNR enhancement by NUS

(\(\eta\))

(Palmer et al, J. Phys. Chem. 2015)
Examples of Theorem 2

Separate cases of Thm 2 could be analytically proven, but notice that we can see the positive slopes and can see visually that these densities have the properties of Thm. 2.
Examples of Theorem 2

Hmmm. Is iSNR increasing?
The Scope of Theorem 2

Exponential Sampling Densities

Gaussian Sampling Densities

The figure shows the ratio of LW_{SMP}/LW_{sig} as a function of time/T_2 for both exponential and Gaussian sampling densities. The graphs depict increasing SNR and match for various sampling densities, including uniform. The figure is based on Palmer et al. (J. Phys. Chem. 2015).
Speculations on Frontiers

- Building consensus on good NUS scheduling
  - Enhancement and line shape are now metrics in identifying good schedules
  - Need more criteria – adherence to weighting, accessibility to broader user base

- What constitutes the ability to detect a peak?
  - Constrained by the raw data prior – iSNR plays a role
  - More to the story

- Vendor Implementations

- Other signal envelopes

- Public Relations.
Consensus on Scheduling

Many recent approaches to schedules (us included) seem to be converging

- Random
- Reproducible / Deterministic
- Minimize Gaps
- Sensitivity and Lineshape
- Ease of Implementation
- Adherence to Weighting
- Generalizable to multiple NUS dimensions
- Generalizable to Partial Component

Propose that the efforts of a number of folks may be converging mathematically.
How to select samples via a density?

- Define the function \( f(x) = (1-x)^2: \ 0 \leq x \leq 1 \).

  The quantile for the function \( f(x) \) divides the function into intervals whose area bounded between two points of an intervals are equal across the function.

  First find the area across \( f(x) \):
  \[
  \int_0^1 (1-x)^2 \, dx = \frac{-(1-x)^3}{3} \bigg|_0^1 = \frac{1}{3}
  \]

  Consider the case of 8 equal quantiles, where there are 7 intervals of equal area across \( f(x) \). Since there are 7 equal intervals, each of the intervals is \( \frac{1}{7} \) of the total area of \( f(x) \).

  Since the function is bounded by 0 and 1, the first and last x value of the quantile is known.

  Using integration we can solve for the second quantile:
  \[
  \int_0^{x_2} (1-x)^2 \, dx = \frac{1}{21} \\
  \frac{-(1-x)^3}{3} \bigg|_0^{x_2} = \frac{1}{21} \\
  (1-x_2)^3 - 1 = \frac{1}{21} \\
  x_2 = 1 - \left(\frac{1}{21}\right)^{\frac{1}{3}}
  \]

  The second quantile is now determined. The third quantile is solved in a similar fashion except now the integration is from 0 to \( x_3 \). This spans two intervals and the area is \( \frac{2}{3} \) or \( \frac{2}{21} \):
  \[
  \int_0^{x_3} (1-x)^2 \, dx = \frac{2}{21} \\
  \frac{-(1-x)^3}{3} \bigg|_0^{x_3} = \frac{2}{21} \\
  (1-x_3)^3 - 1 = \frac{2}{21} \\
  x_3 = 1 - \left(\frac{2}{21}\right)^{\frac{1}{3}}
  \]

  The general expression for 8 quantiles:
  Let \( z = \int_0^{x_8} (1-x)^2 \, dx \), where \( x_1 = 0 \) and \( x_8 = 1 \).

  Each of the 8 quantiles will be represented as elements of the 1x8 vector \( a \):
  \[
  a_j = \left(\frac{j-1}{n}\right) \int_0^{x_j} (1-x)^2 \, dx - z
  \]

  This can be manipulated for the general case of n quantiles:
  Let \( z = \int_0^{x_n} (1-x)^2 \, dx \), where \( x_1 = 0 \) and \( x_n = 1 \).

  Each of the n quantiles will be represented as elements of the n-dimensional vector \( a \):
  \[
  a_j = \left(\frac{n-1}{j}\right) \int_0^{x_j} (1-x)^2 \, dx - z
  \]

- Random
- Reproducible / Deterministic
- Minimize Gaps
- Sensitivity and Lineshape
- Ease of Implementation
- Adherence to Weighting
- And more…

(ms in preparation; if you use please reference Private Communication with DSR and VGR)
How to select samples via a density?

(ms in preparation; if you use please reference
Private Communication with DSR and VGR)
Vendor implementations*

Uniformly organize NUS data in memory; NUS schedule always in header.

More transparent/user-enabled approach to weighting.

Automate NUS from oversampled grid.

Automate real-time and time-resolved NUS.

Calculators to estimate $T_2^*$ and to estimate enhancements.

Need a suite of standardized test data sets so that schedules/processing can be certified.

*Some of these may already be addressed.
Other Signal Envelopes

Fig. 10. Comparison of (a) the matched DQFC experiment ($t_1 = 11-59$ ms) with (b) the normal DQFC experiment ($t_1 = 0-48$ ms) for an expanded spectral region. The experiment in (a) was done as described in the legend to Fig. 9. Both spectra have an identical number of total accumulations.


Peter Schmieder, Alan S. Stern, Gerhard Wagner, Jeffrey C. Hoch, “Application of nonlinear sampling schemes to COSY-type spectra”,

P-Set-3.3
Starting to see more formal assurances for NUS from several avenues. Important to increase breadth of understanding of these fundamentals.

- Improved understanding/reassurance of spectral windows.

- Improved/expanded metrics for schedules.

- More routine reports on NUS in two or more indirect dimensions.

- Improved understanding of sensitivity, with strong theorems.

Need to be cautious to distinguish bleeding edge applications from established, robust NUS applications, and cautious about overuse of the straw man.
• Exact numeric enhancement
• Compound in multiple dimensions.
• Compound with any hardware-derived enhancements.
• Guesswork eliminated - allows for rational design at detection limits