Interferogram and real-time acquisition methods

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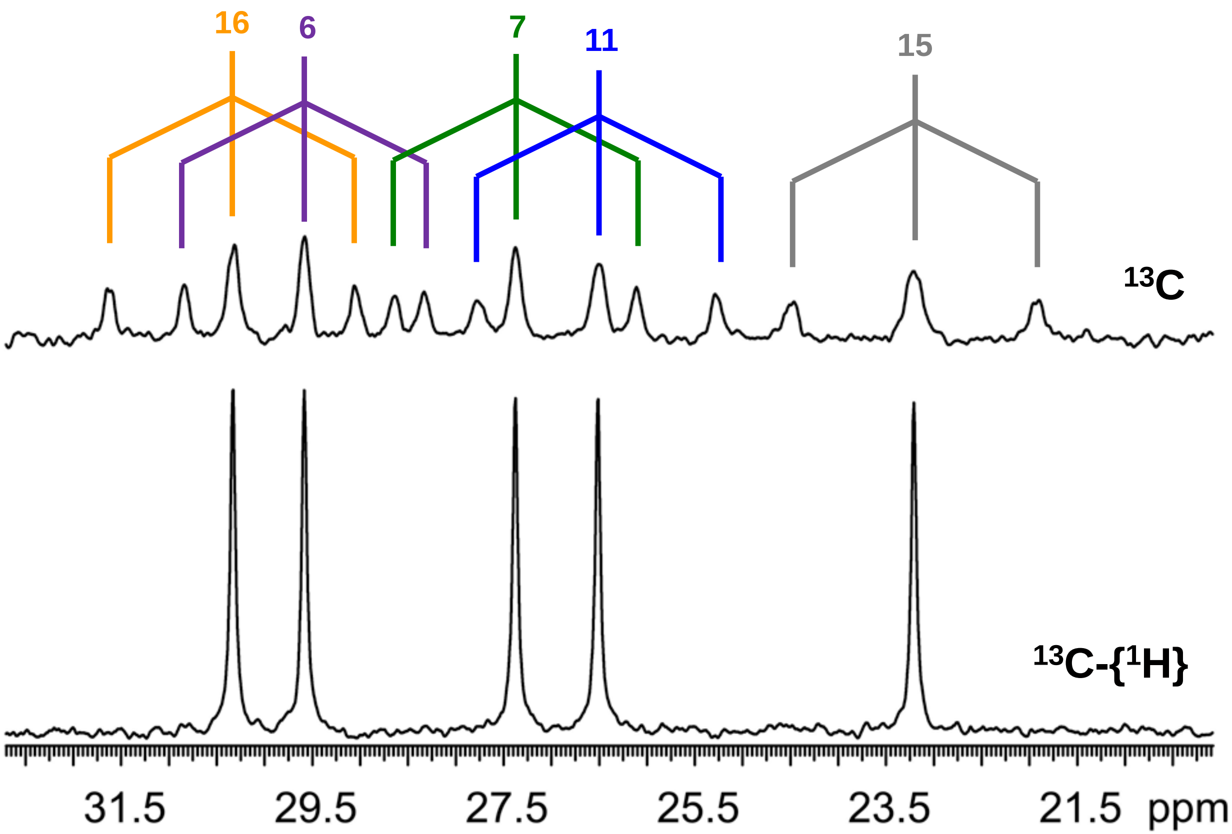
Workshop on Pure Shift NMR

12th Sep 2017

Outline

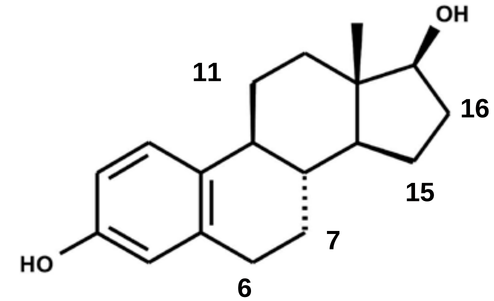
- Why are we interested in decoupling?
- Basic concepts in pure shift NMR
  - Active and passive spins
  - Active spin refocusing elements: BS / Zangger-Sterk / PSYCHE / BIRD
- The interferogram acquisition method
  - Theory and pulse sequences
  - Illustrative examples
- The real-time acquisition method
  - Theory and pulse sequences
  - Illustrative examples
- Summary

Suppressing multiplet structure – heteronuclear decoupling



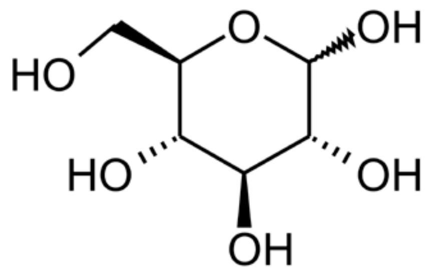
proton
coupled

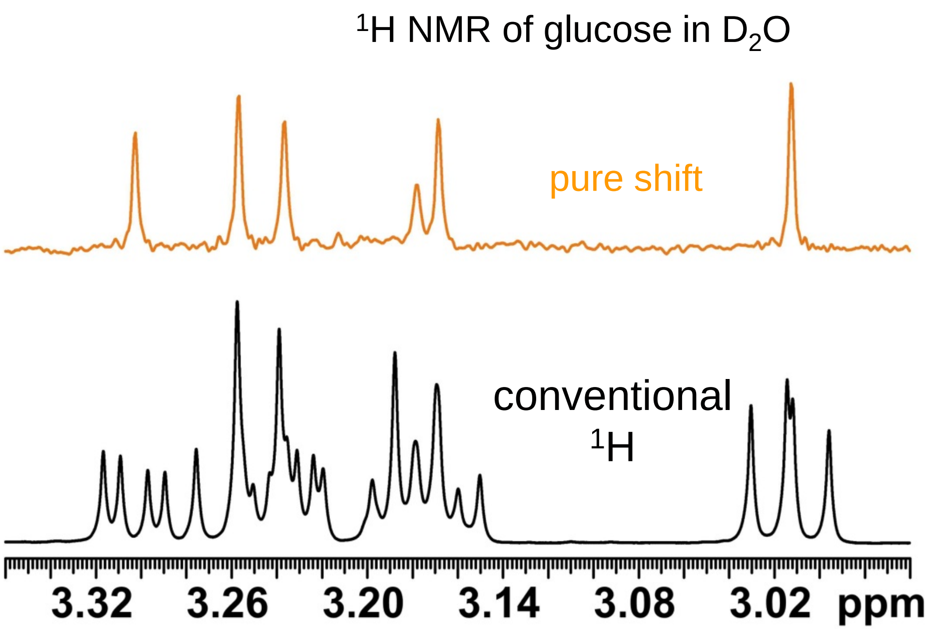
proton
decoupled



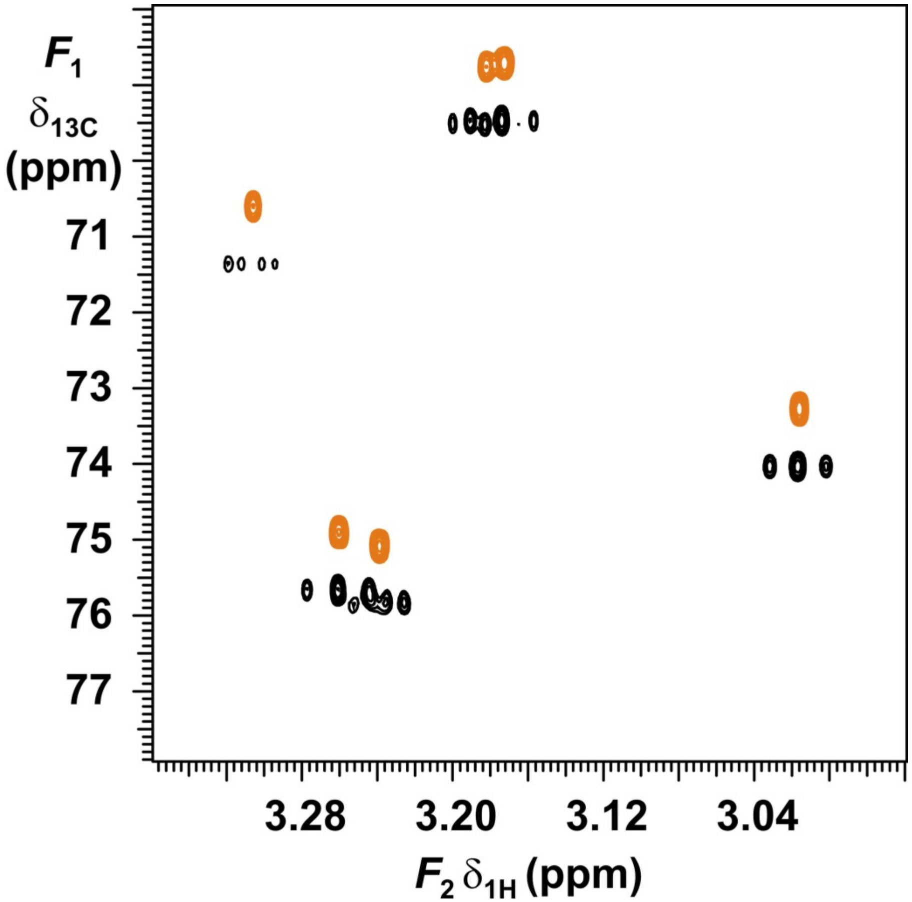
Partial ^{13}C NMR spectrum of estradiol in DMSO-d$_6$ with (bottom) and without (top) proton decoupling

Suppressing multiplet structure – homonuclear decoupling

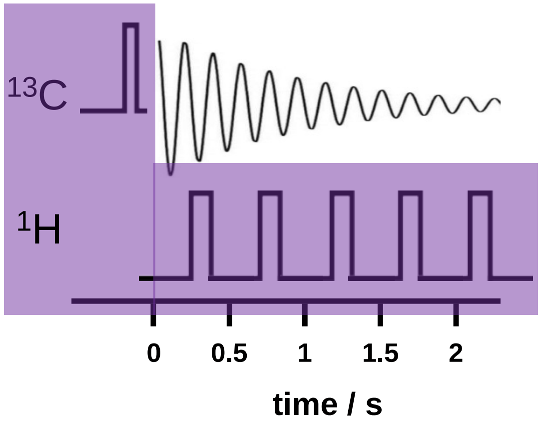




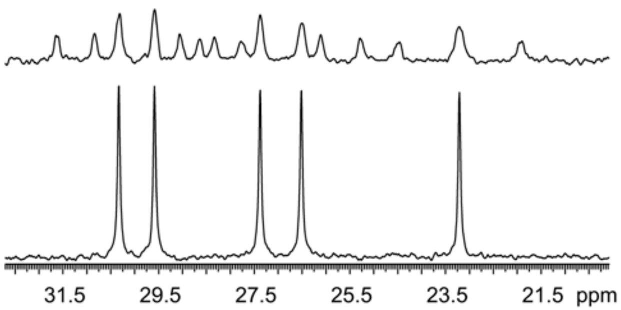
Partial ^{1}H NMR spectrum of glucose in D_2O

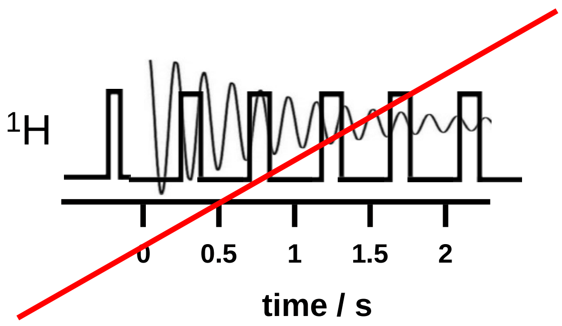


The concepts of spin decoupling



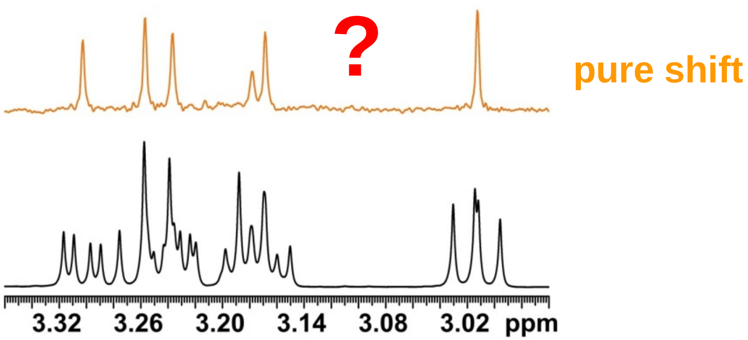
Heteronuclear decoupling
WALTZ / GARP / adiabatic bi-level



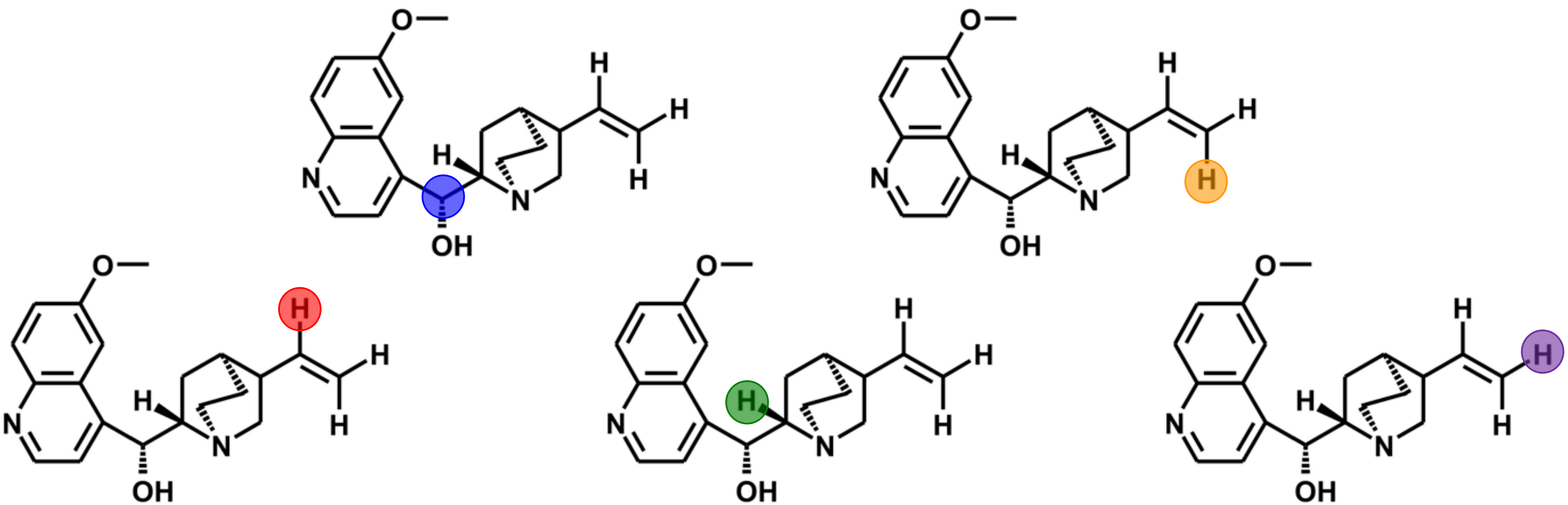


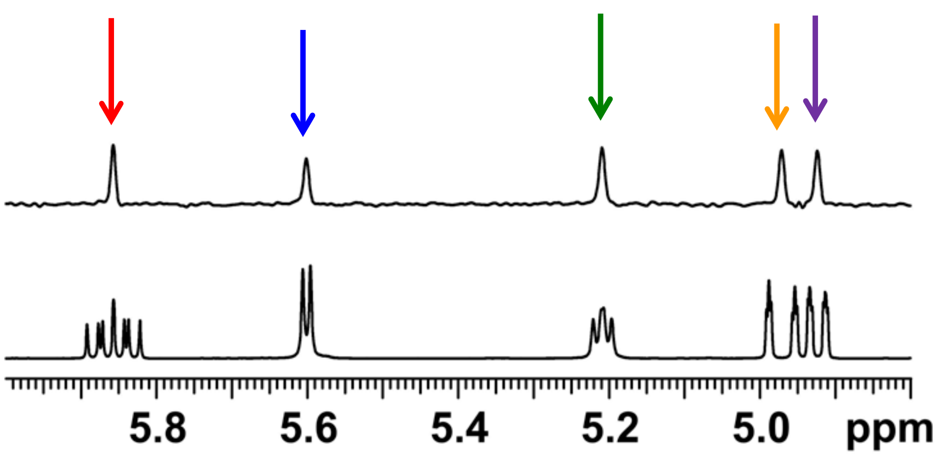
Homonuclear decoupling

1. **time management**
2. **selectivity problem**



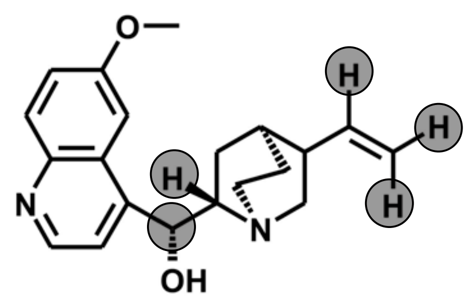
Active and passive spins in pure shift NMR





pure shift

conventional
^{1}H



50 mM quinine in dmso-d_6

ASR - Active Spin Refocusing - elements

Magn. Reson. Chem. **35**, 9 (1997)

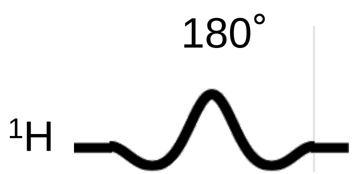
J. Magn. Reson. **124**, 486 (1997)

Angew. Chem. Int. Ed. **53**, 6990 (2014)

Chem. Phys. Lett. **93**, 504 (1982)

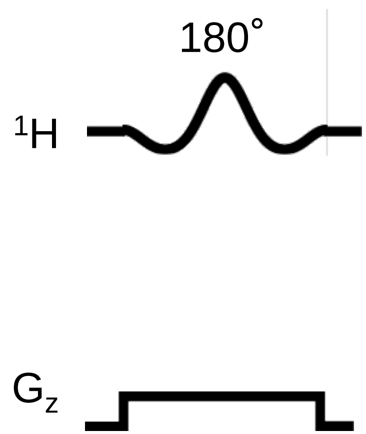
BS

frequency selective RSNOB pulse



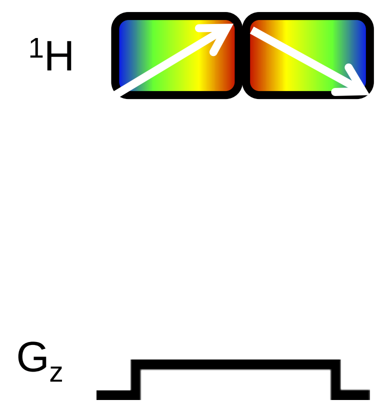
Zangger-Sterk

frequency selective RSNOB pulse

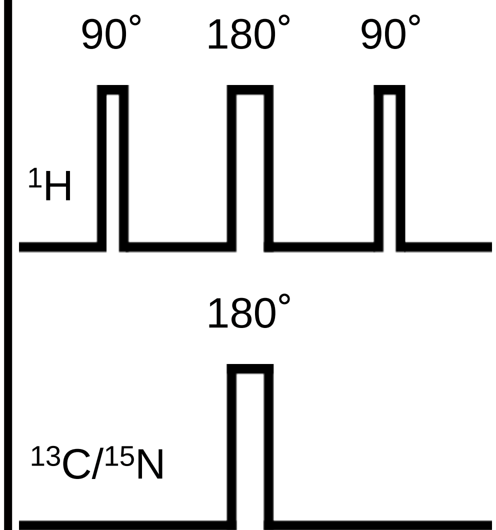


PSYCHE

Small flip angle CHIRP pulses



BIRD



all work by differentially manipulating **active** and **passive** subpopulations of protons

ASR - Active Spin Refocusing - elements

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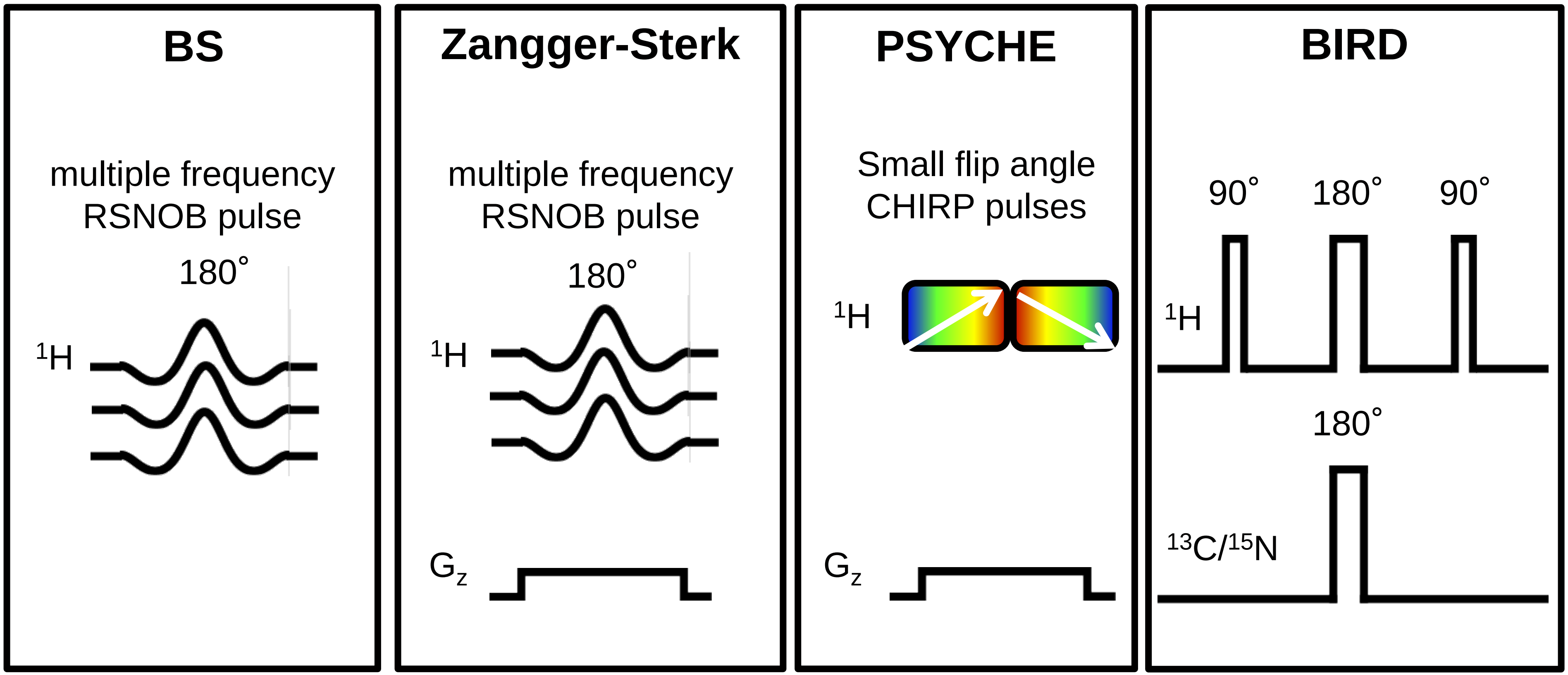
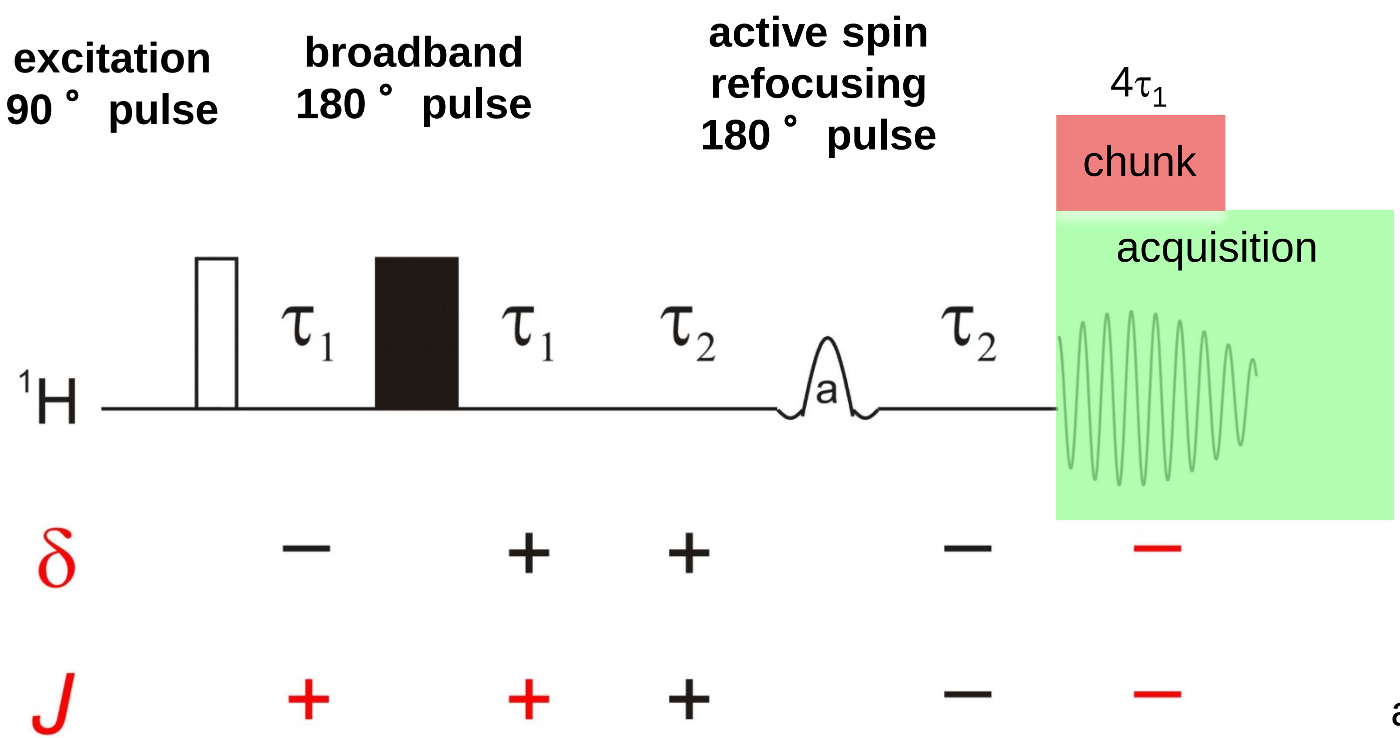


all work by differentially manipulating **active** and **passive** subpopulations of protons

Double spin echo experiment with an ASR element



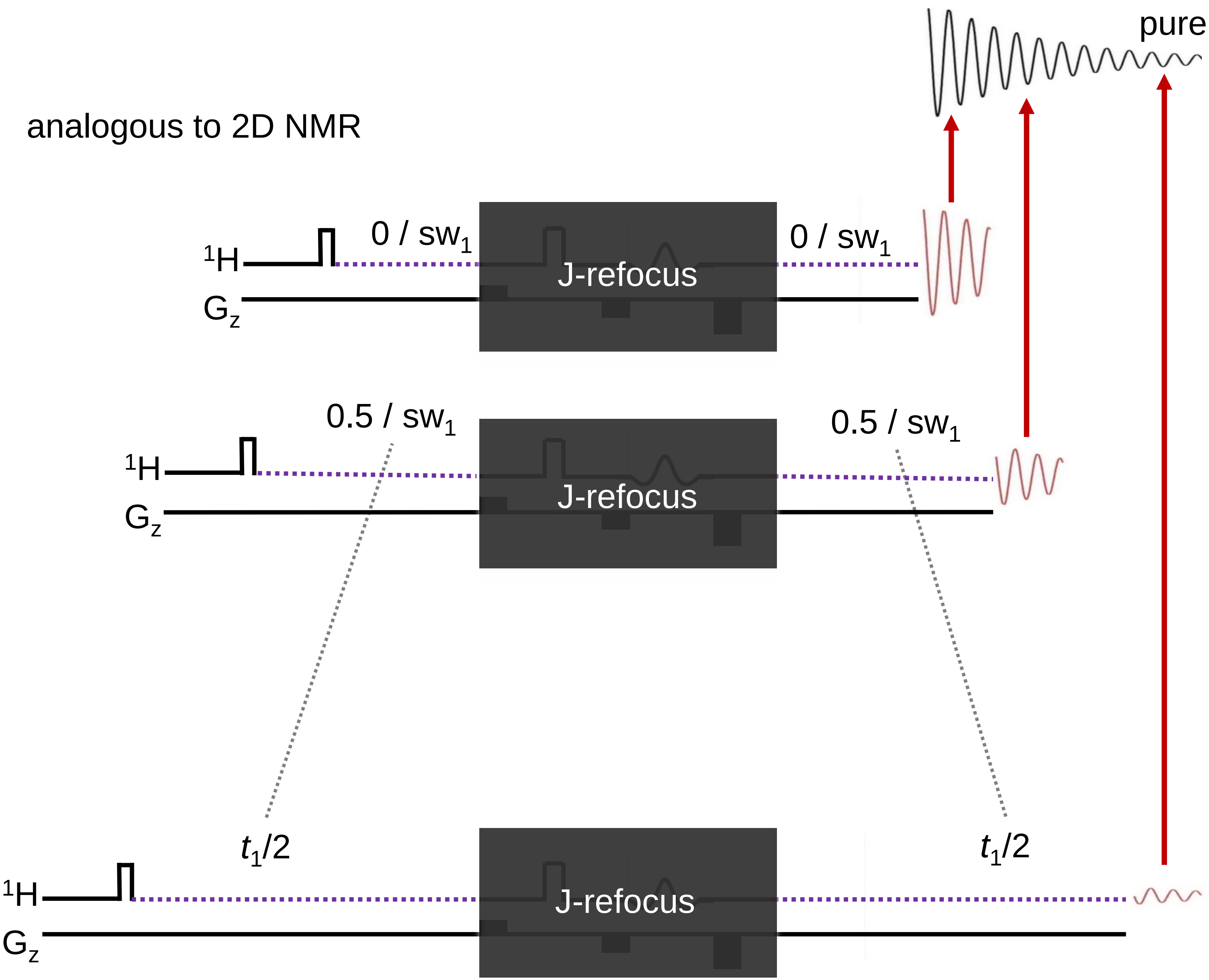
chemical shift is refocused at the beginning of acquisition, but …

… homonuclear coupling (J) is refocused $2\tau_1$ later

J-evolution is negligible during the first few data points (period $4\tau_1$) delivering a small chunk of the desired pure shift FID

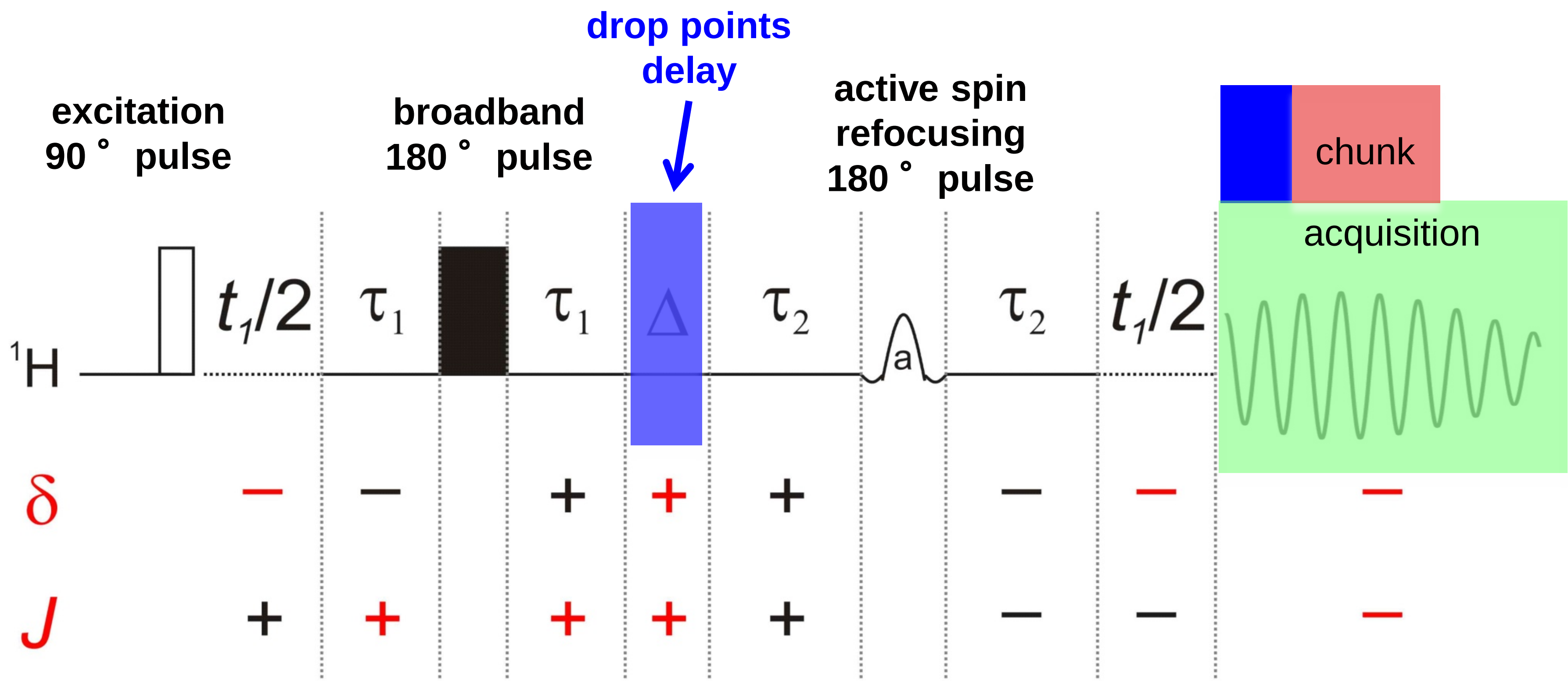
Interferogram pure shift experiment – 2D acquisition

analogous to 2D NMR



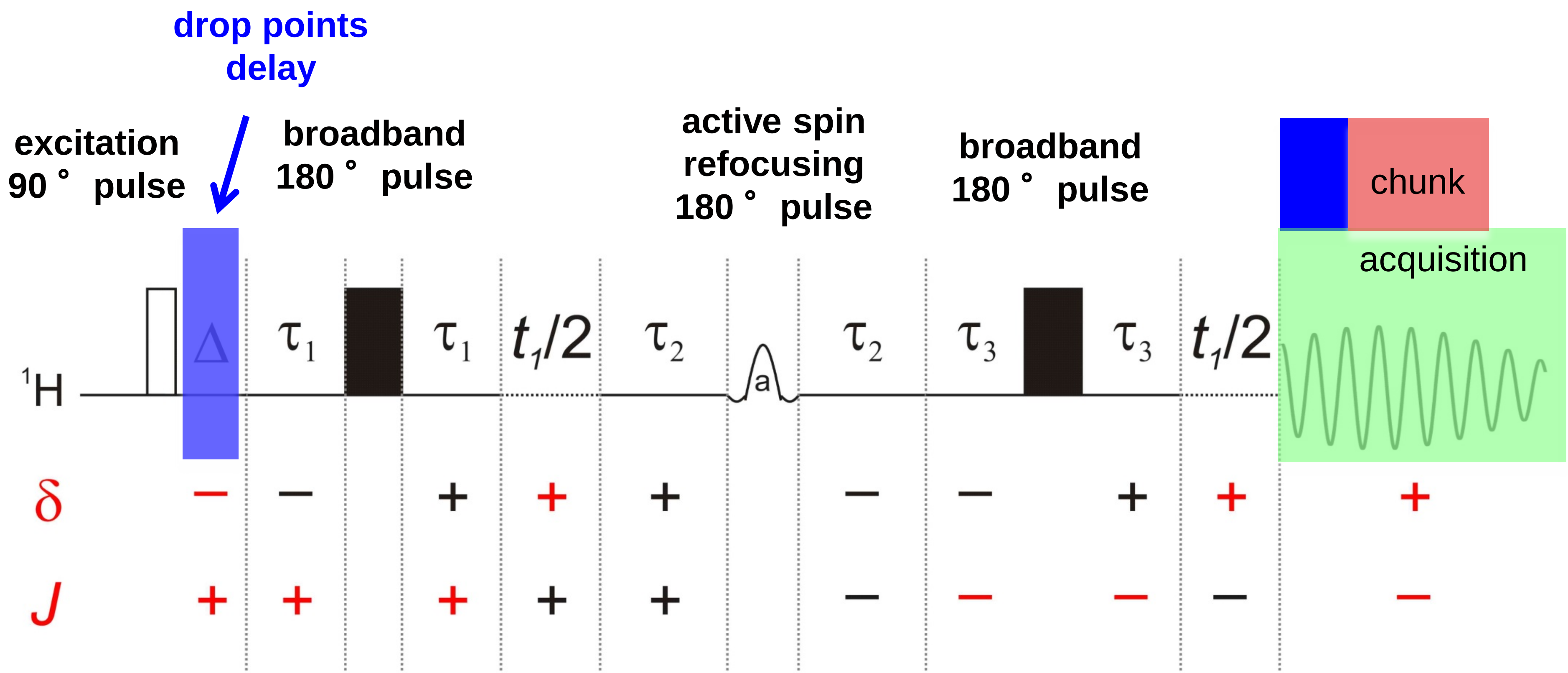
$J \ll \delta$ allows data acquisition in 'chunks' of duration $1/sw_1$

The interferogram pure shift pulse sequence



- Chemical shift is refocused Δ = [*integer*] / sw later than the beginning of acquisition
- *J* is refocused at the midpoint of each chunk ($4\tau_1$)
- Incremented delay allows recording all chunks of pure shift FID (during t_1 δ evolves, but *J* is refocused)
- Phase cycle and/or PFG pairs can enforce CTP selection

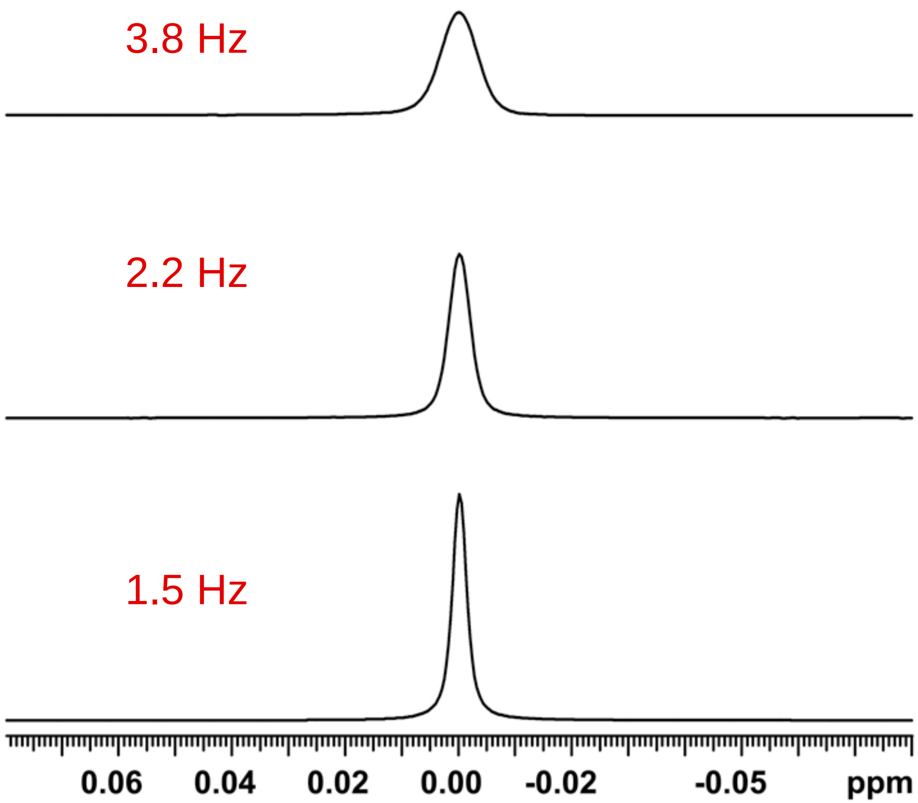
The triple spin echo analogue with an ASR element



- Frequency-swept broadband pulses can be implemented
- Useful to deal with strong coupling artefacts e.g. in TSE-PSYCHE
- Duration of the chunk is not limited by duration of gradient pulses, but more relaxation loss

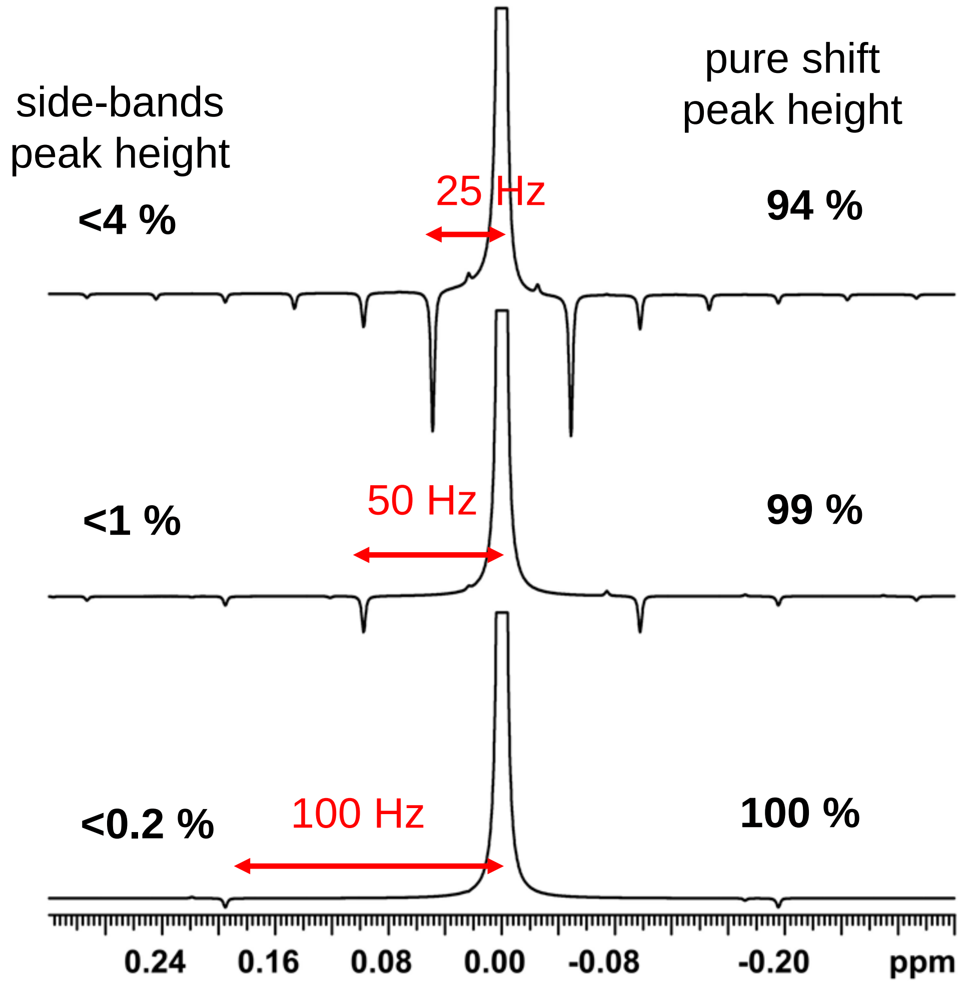
Resolution in interferogram pure shift experiments

line-width at half height	number of chunks	duration of the complete interferogram (pure shift acquisition time)
3.8 Hz	16	0.3 s
2.2 Hz	32	0.6 s
1.5 Hz	64	1.2 s



Interferogram band-selective pure shift spectra of an AX spin system calculated in Matlab/Spinach. (20 ms chunk duration; J_{AX} = 10 Hz)

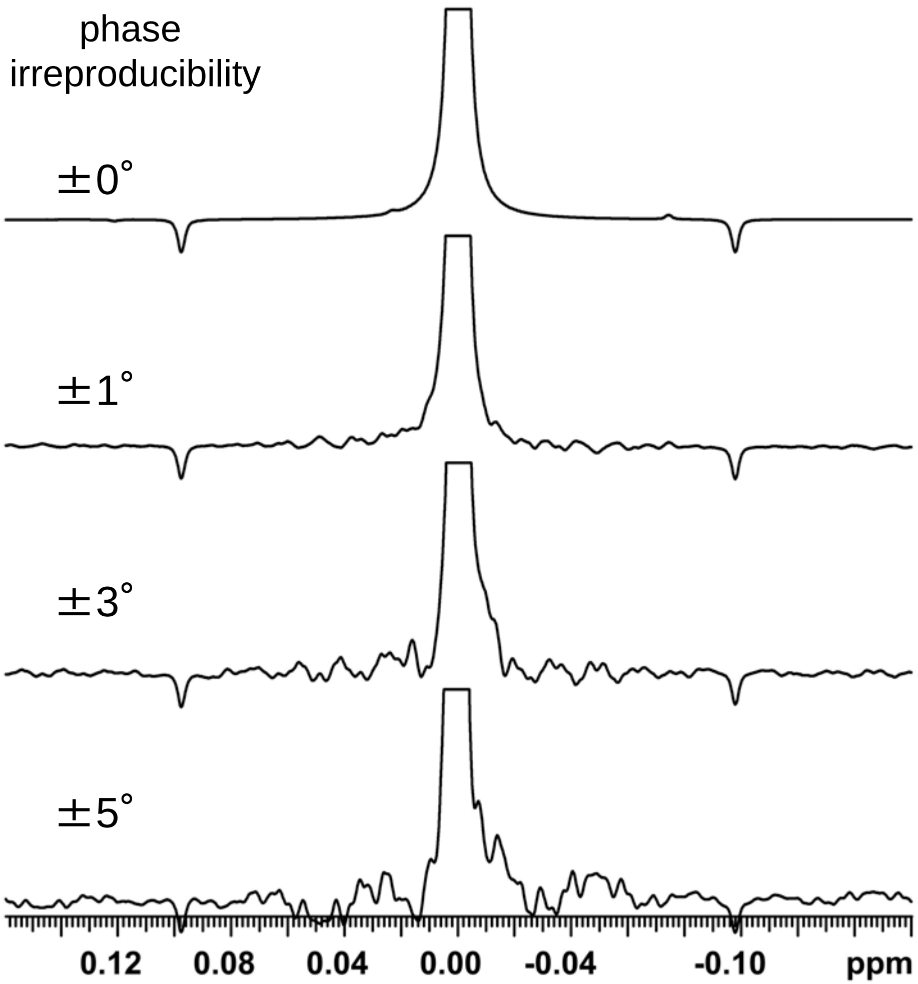
Effect of duration of chunk (SW_1)



duration of chunk $1/SW_1$ (SW_1)	number of chunks (TD1)
40 ms (25 Hz)	32
20 ms (50 Hz)	64
10 ms (100 Hz)	128

Interferogram band-selective pure shift spectra of an AX spin system calculated in Matlab/Spinach. (AQ = 1.3 s; J_{AX} = 10 Hz)

Effect of RF phase instability (scan-to-scan)



Interferogram band-selective pure shift spectra of an AX spin system calculated in Matlab/Spinach. (AQ = 1.3 s; J_{AX} = 10 Hz; SW_1 = 50 Hz)

analogous to t_1-noise in 2D NMR

interleaved
2D data acquisition
is advisable for long
experiments

Speeding things up

interferogram approach

J. Magn. Reson.
124, 486 (1997)

real-time approach

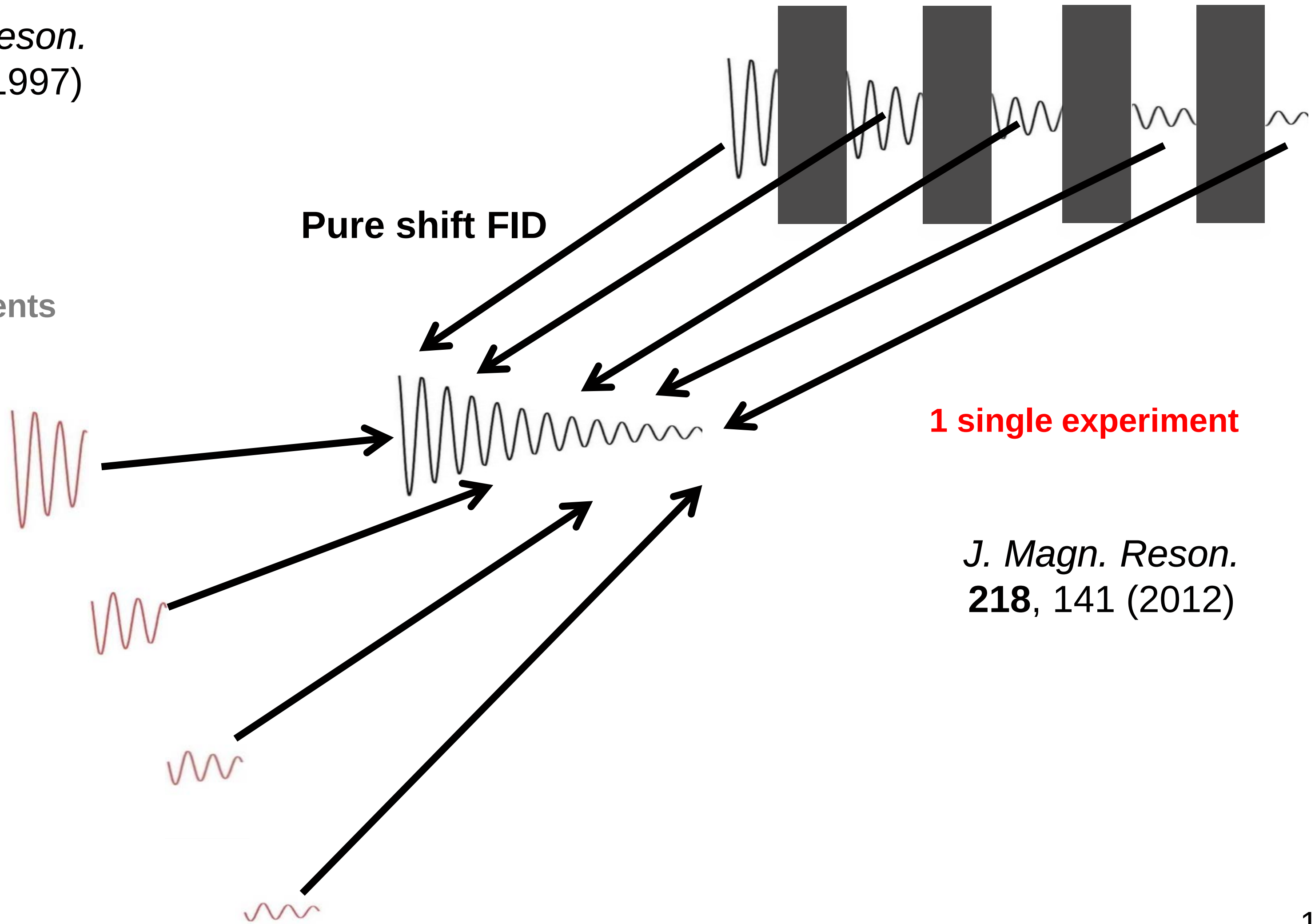
a set of experiments

1st experiment

2nd experiment

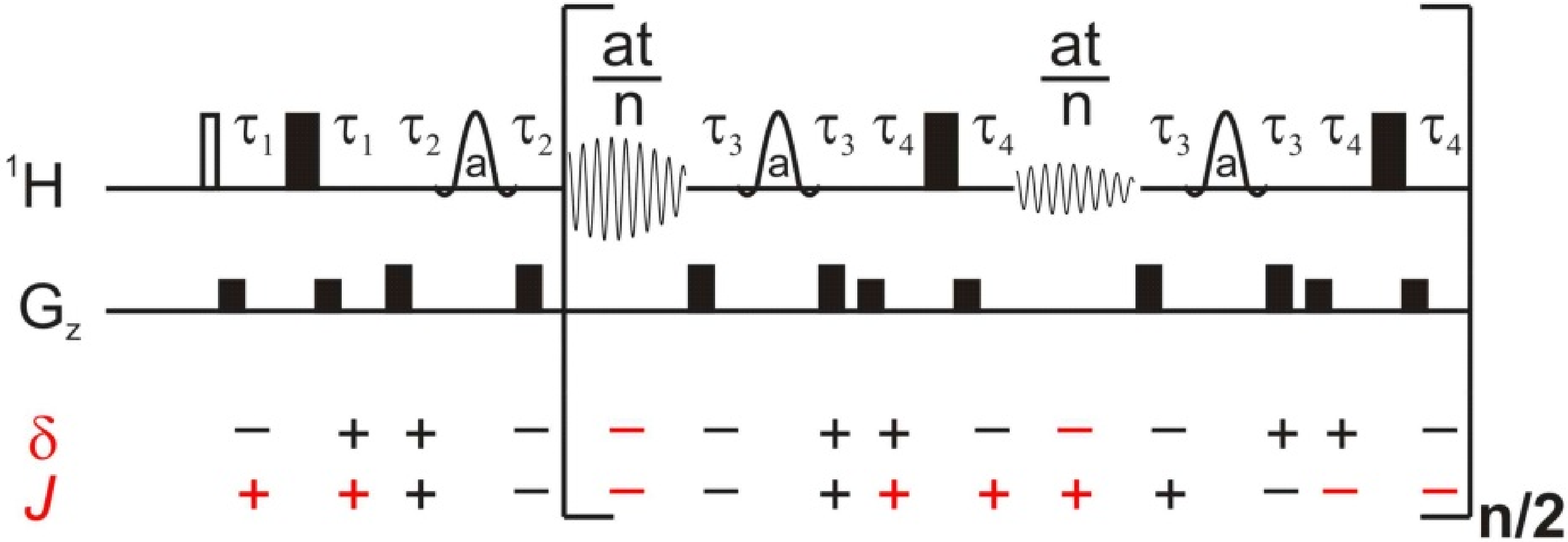
3rd experiment

4th experiment



J. Magn. Reson.
218, 141 (2012)

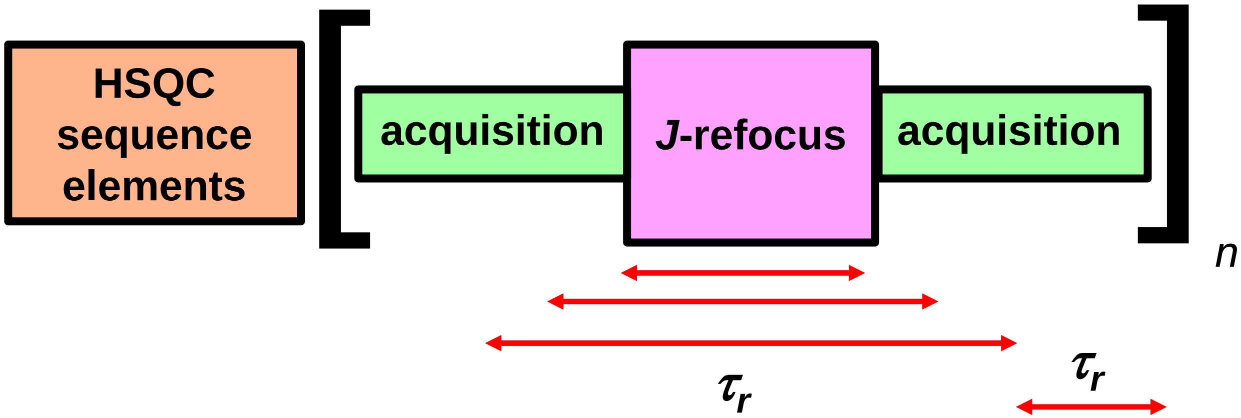
Prototype pulse sequence of real-time pure shift NMR



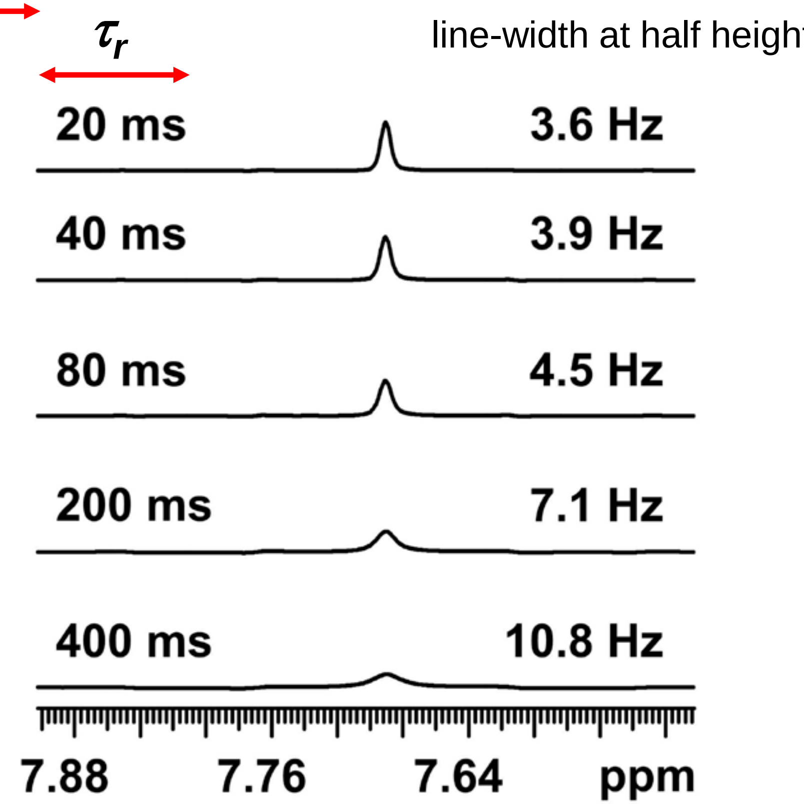
- Chunks are collected from a single scan
- Resolution is worse compared to interferogram experiment, because of

  relaxation, diffusion/convection, and cumulative errors of pulse imperfection

Relaxation losses in real-time experiments

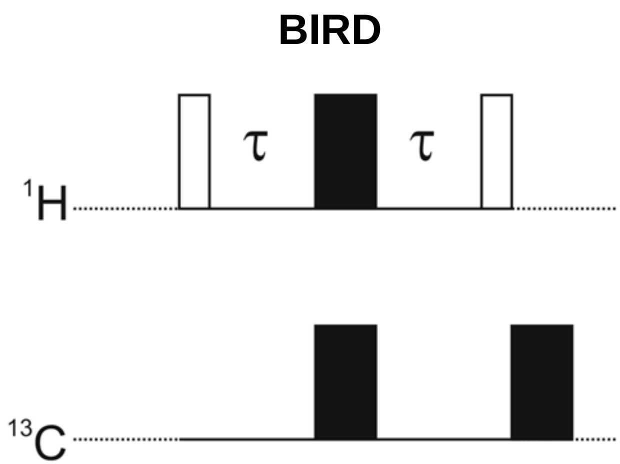


Selected traces of real-time pure shift HSQC BIRD.
The total echo time (τ_r) of the *J*-refocusing element was incremented.



Broadening here is due to proton T_2 relaxation.

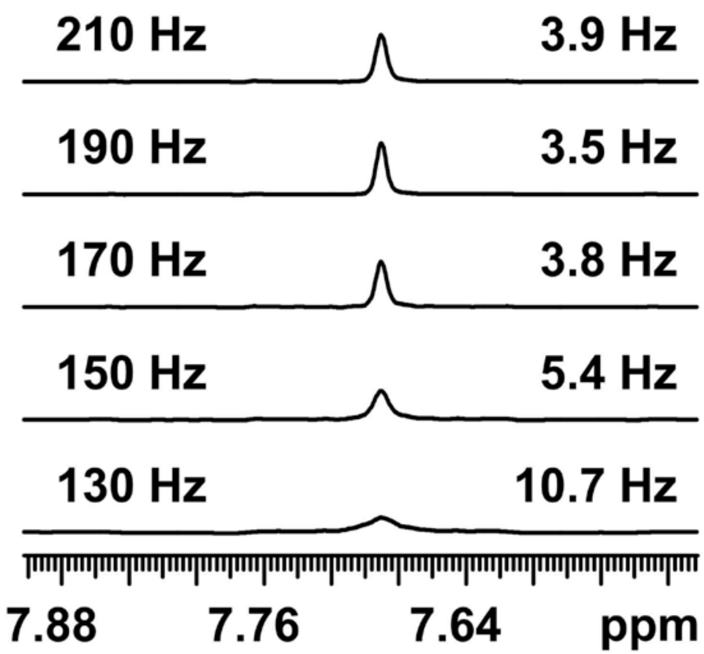
Effect of BIRD timing error



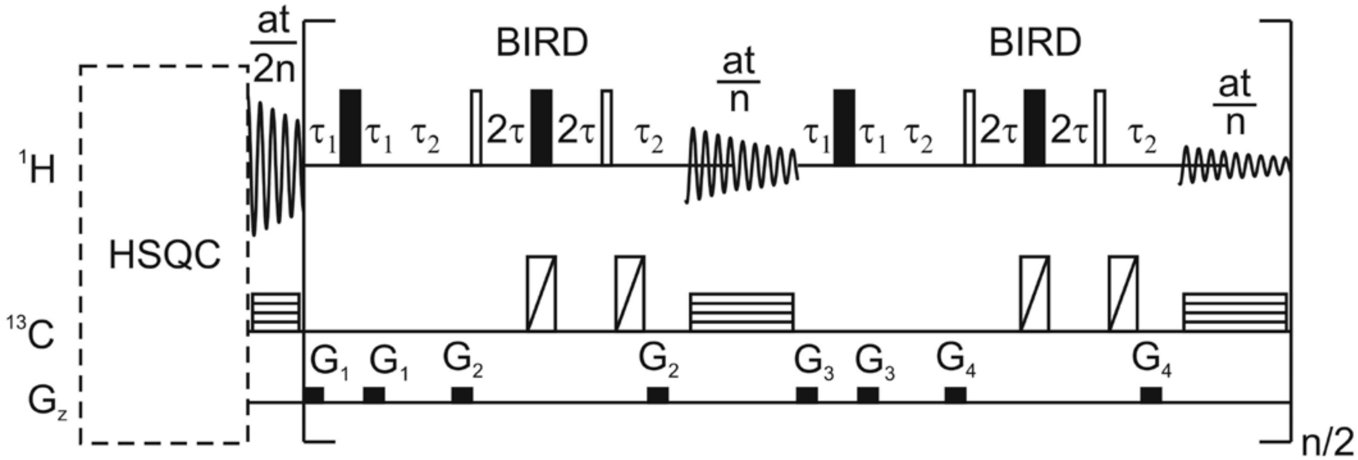
If $\tau = 1 / (2 * {}^1J_{CH})$, then BIRD element refocuses the ^{13}C-attached protons.

Selected traces of real-time pure shift HSQC BIRD (^{1}CH = 190 Hz).
The echo time (τ) of the BIRD element was varied (HSQC sequence element was kept constant).

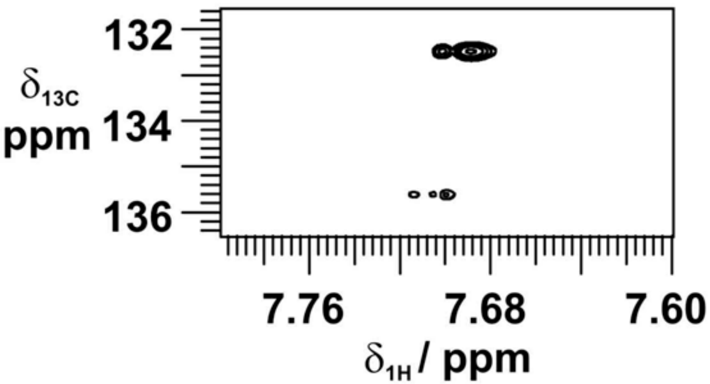
Broadening here is due to BIRD timing error.



Effect of pulse imperfection in real-time pure shift HSQC using BIRD

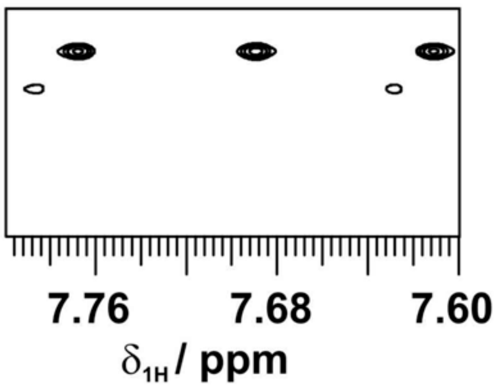


Gradient pulses omitted, basic phase cycle, and no supercycle…



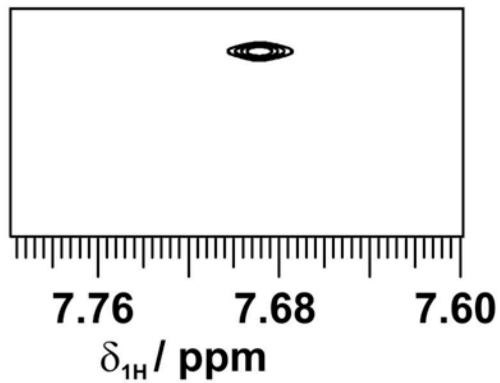
… extra signals next to the pure shift signal and F_1 mirror images

2nd carbon 180 pulse is omitted in BIRD…



… extra signals appear due to heteronuclear *J*-evolution during τ_2

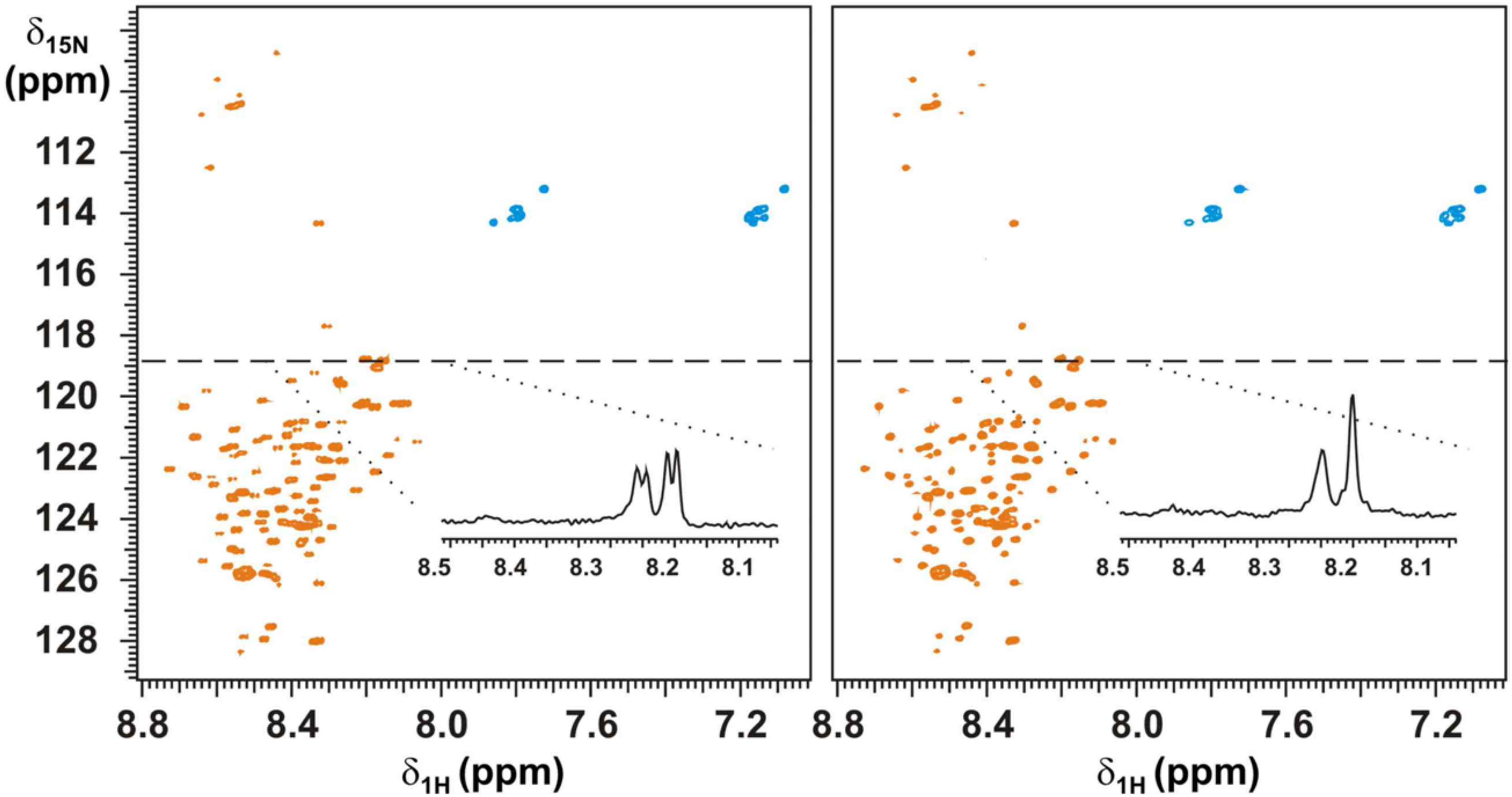
$G_{1\text{-}4}$ varied chunk-to-chunk extended phase cycle (*2), and MLEV-16 supercycle…

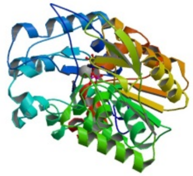


… clean

^{15}N HSQC of L80C mutant N-PGK protein in water (90%H_2O / 10%D_2O)

^{15}N HSQC | pure shift ^{15}N HSQC

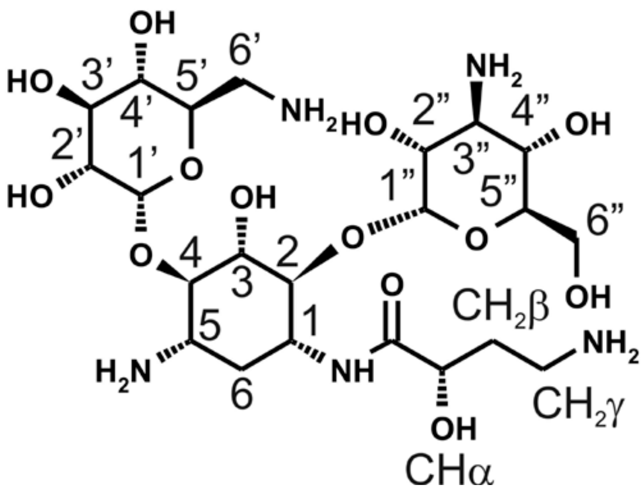


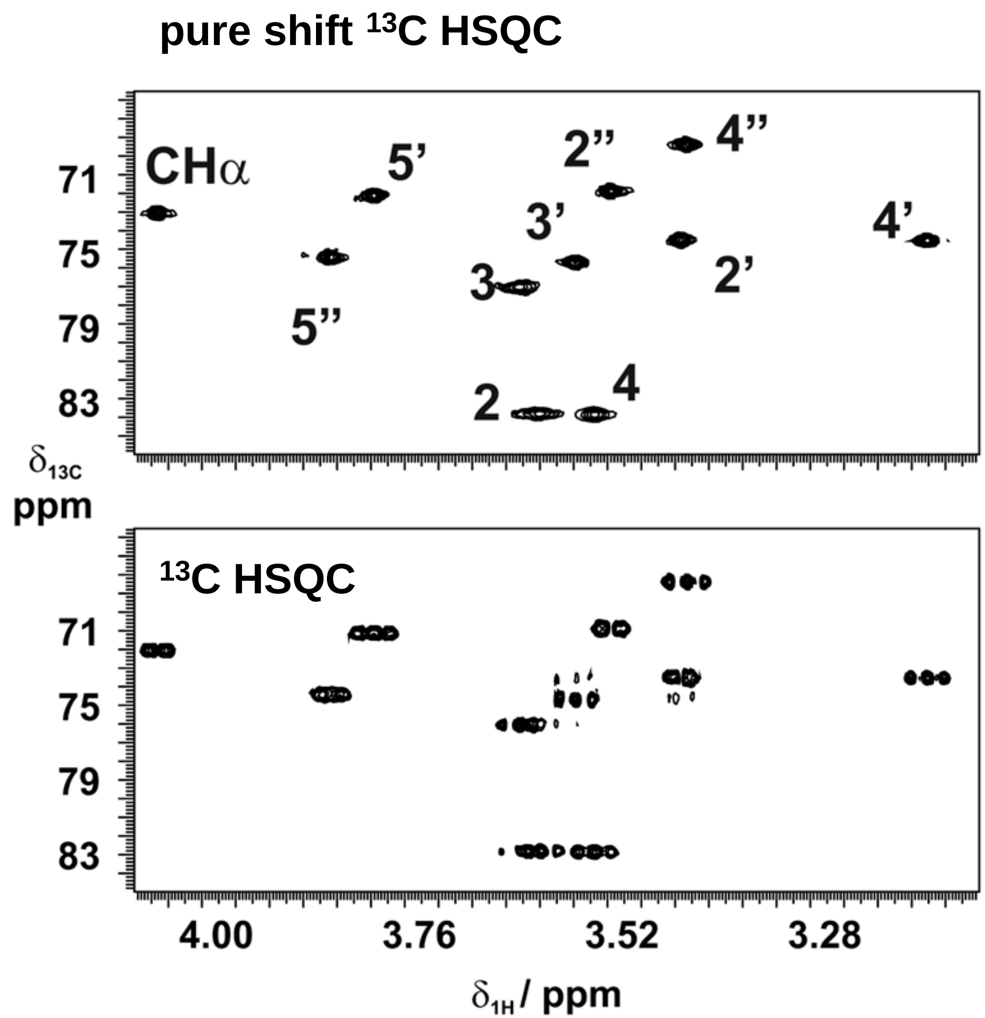


N-terminal domain of PGK

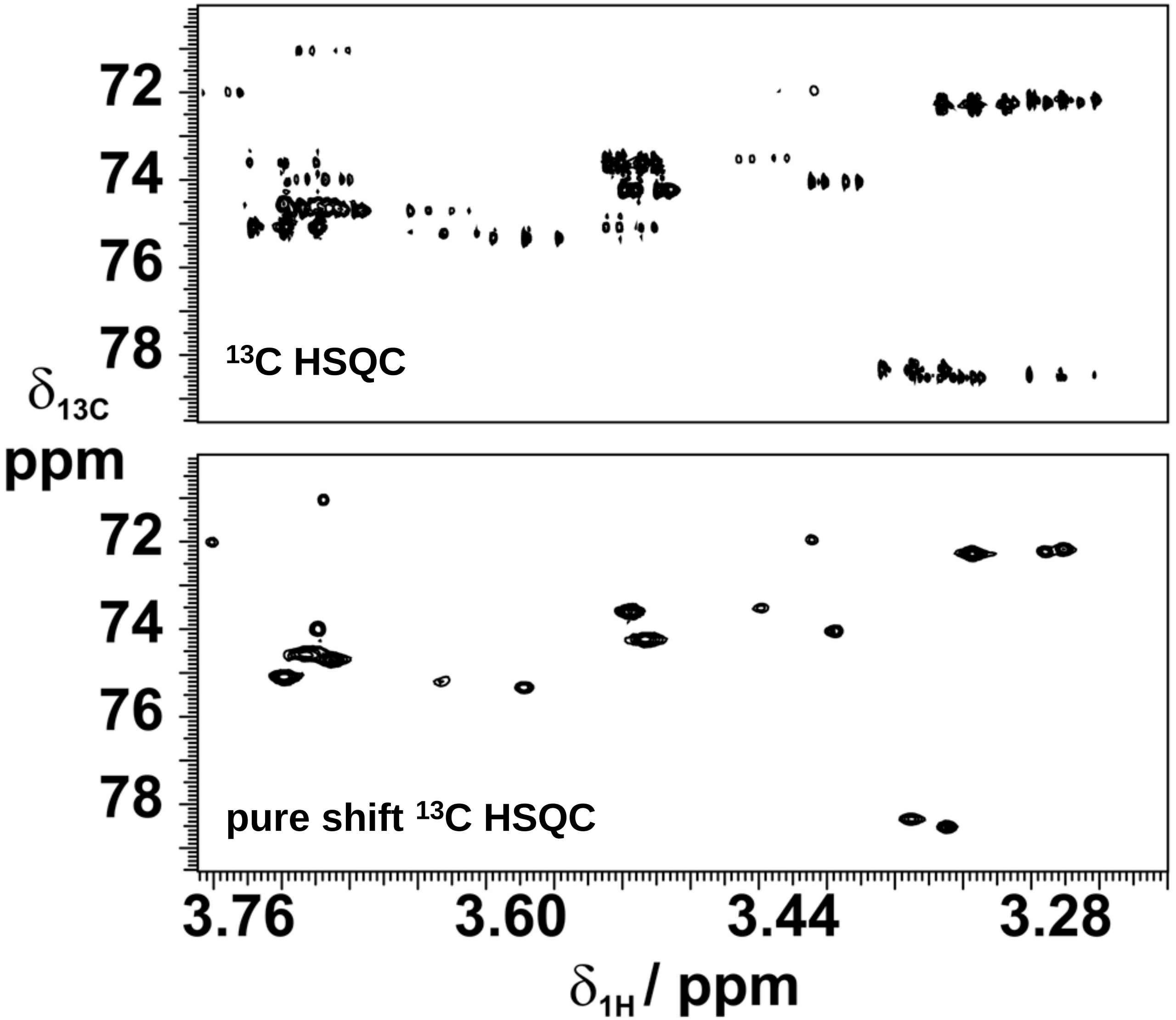
J. Biomol. NMR. **62**, 43 (2015)

10 mg amikacin in D_2O

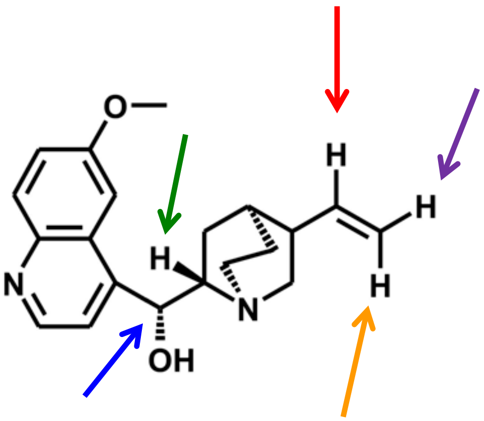


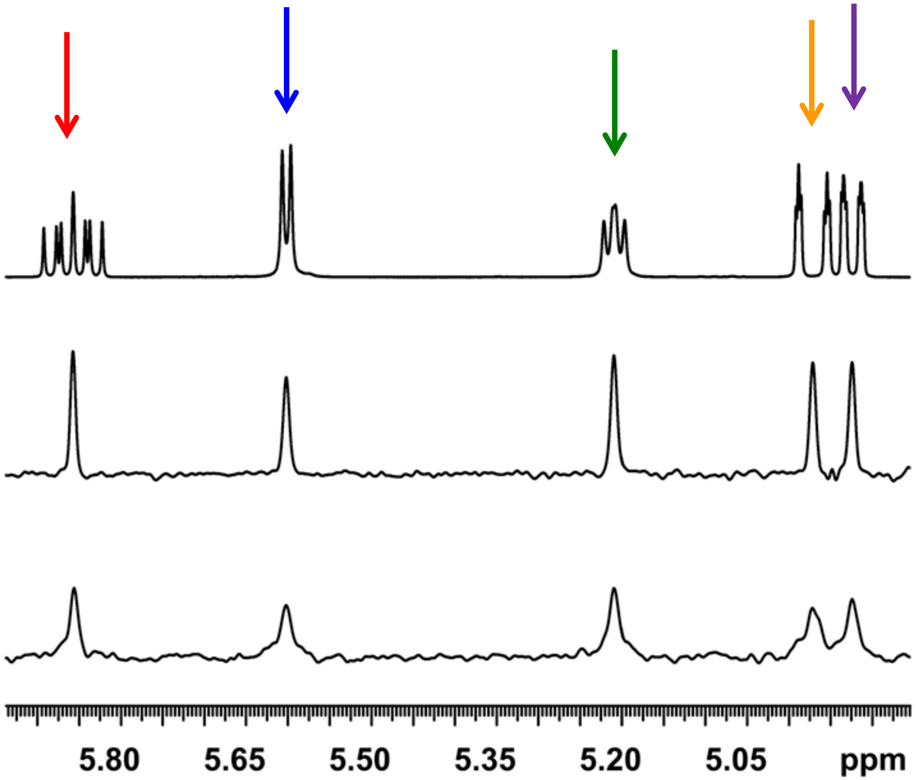


Mixture of glucose, trehalose, raffinose, and α-cyclodextrin



Interferogram vs real-time pure shift NMR





experiment time

conventional
^{1}H — 0.5 min

interferogram
Zangger-Sterk — 5 min 30 s

real-time
Zangger-Sterk — 0.5 min

50 mM quinine in dmso-d_6

Summary

Interferogram experiments

- Resolution enhancement requires extended experiment time
- There is a trade-off between experiment time and quality of spectrum
- Many kinds of active spin refocusing element are available
- System instability may affect very high resolution experiments

Real-time acquisition

- Experiment times of parent and pure shift methods are practically identical
- Simultaneous sensitivity and resolution enhancement in real-time HSQC using BIRD
- Resolution enhancement is limited by relaxation, diffusion/convection, and pulse imperfection
- Lock disturbance can be a problem in high resolution experiments

Acknowledgements

Gareth Morris, Mathias Nilsson

and the NMR Methodology Group

http://nmr.chemistry.manchester.ac.uk/

Funding

EMBO

MARIE CURIE FELLOWSHIPS

Marie Curie Fellowships

EPSRC

Engineering and Physical Sciences Research Council

A Pure Shift NMR Workshop

11.00	Gareth Morris	**Welcome, introduction and history**
11.30	Peter Kiraly	**Interferogram and real-time acquisition methods**
12.00	Laura Castañar	**Zangger-Sterk and band-selective methods**
12.30	Mohammadali Foroozandeh	**PSYCHE**
13.00		*Lunch and poster session*
14.00	Ralph Adams	**Other pure shift and related methods**
14.30	Mathias Nilsson	**Practical implementations**
15.00	Adolfo Botana	**JEOL pure shift implementation**
15.10	Vadim Zorin	**MestreNova pure shift implementation**
15.20	Ēriks Kupče	**Bruker shaped pulse implementation**
15.30		*Question and answer session*

University of Manchester, 12th September 2017