

Supporting Information for

Spin dynamics of [$^{13}\text{C}_2$]pyruvate hyperpolarization by parahydrogen in reversible exchange at micro Tesla fields

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Table of Contents

1) Full 4-Spin Level Anti Crossing Plot of [1,2- $^{13}\text{C}_2$]Pyruvate	2
2) First Derivative of SF1	2
3) Lifetime of $^{13}\text{C}_2$ Pyruvate at Varied Fields.....	3
4) 4 Spin Matrix of [1,2- $^{13}\text{C}_2$]Pyruvate	5
5) MATLAB Simulation	6
6) Individual Spin State Contribution	10

1) Full 4-Spin Level Anti Crossing Plot of [1,2-¹³C₂]Pyruvate

The full LAC plot showing all spin state transitions for [1,2-¹³C₂]pyruvate was calculated and is shown below. A portion of this plot was shown in the main text to highlight the specific transitions that were being studied, but it brings a more complete picture to show the entire LAC plot here. The legend on the right color codes each initial spin state and each possible transition can be identified by avoided crossings throughout the increase in field strength from left to right.

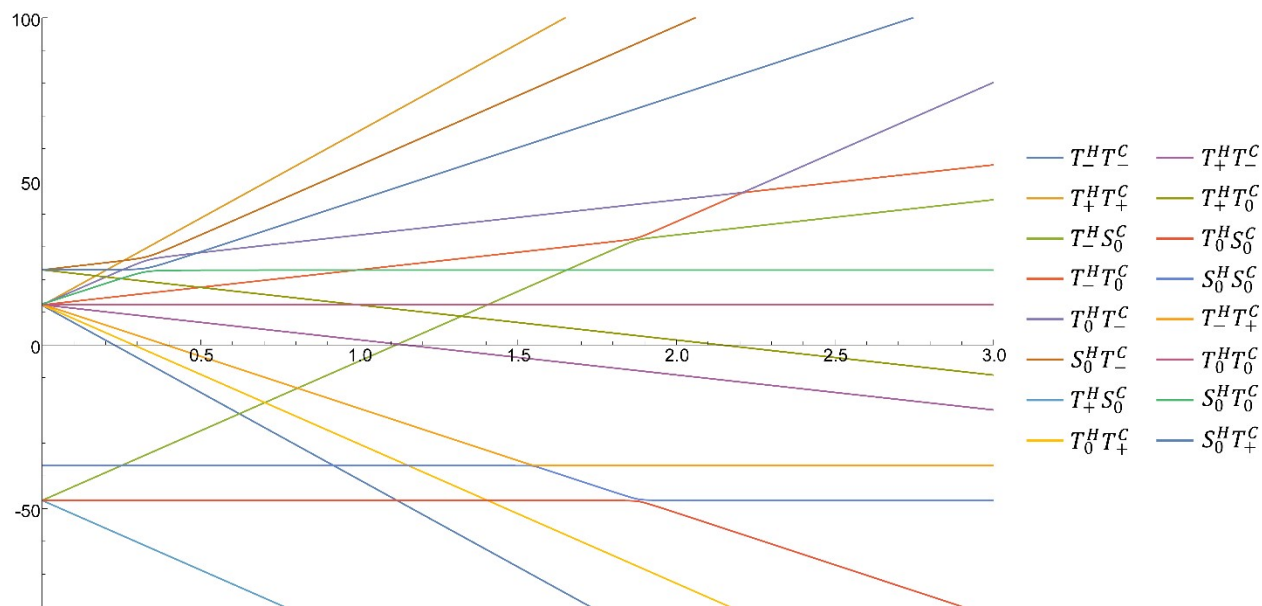


Figure S1. The full LAC plot with all interactions between spin states in the [1,2-¹³C₂]pyruvate system from 0 to 3 μT.

2) First Derivative of SF1

The LAC plot was further analyzed by taking the first derivative to extrapolate the rate at which the spin states transition. This figure shows another useful tool for identifying avoided crossings and diving into the dynamics behind them.

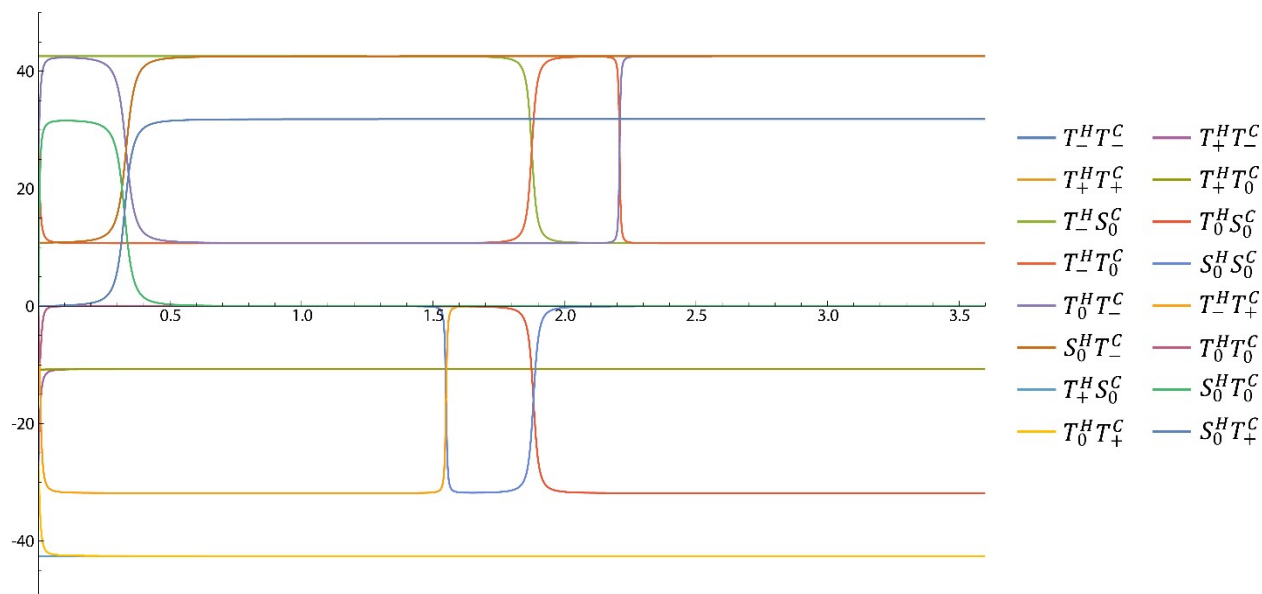


Figure S2. Shown here is the first derivative of S1 which shows the rate at which spin energy change as a function of magnetic field.

3) Lifetime of $^{13}\text{C}_2$ Pyruvate at Varied Fields

Here we show how the lifetime data was fitted to calculate the T_1 or T_s . As discussed in the main paper after 0.17 T the singlet state is no longer an eigenstate. This lead to not only a data collection of T_s data by taking the difference in the C1 and C2 peaks and looking at the decay of the difference, but also a collection of T_1 data on both carbon 1 of $[1,2-^{13}\text{C}_2]$ pyruvate and carbon 2 of $[1,2-^{13}\text{C}_2]$ pyruvate by looking at the individual peak decays at each field.

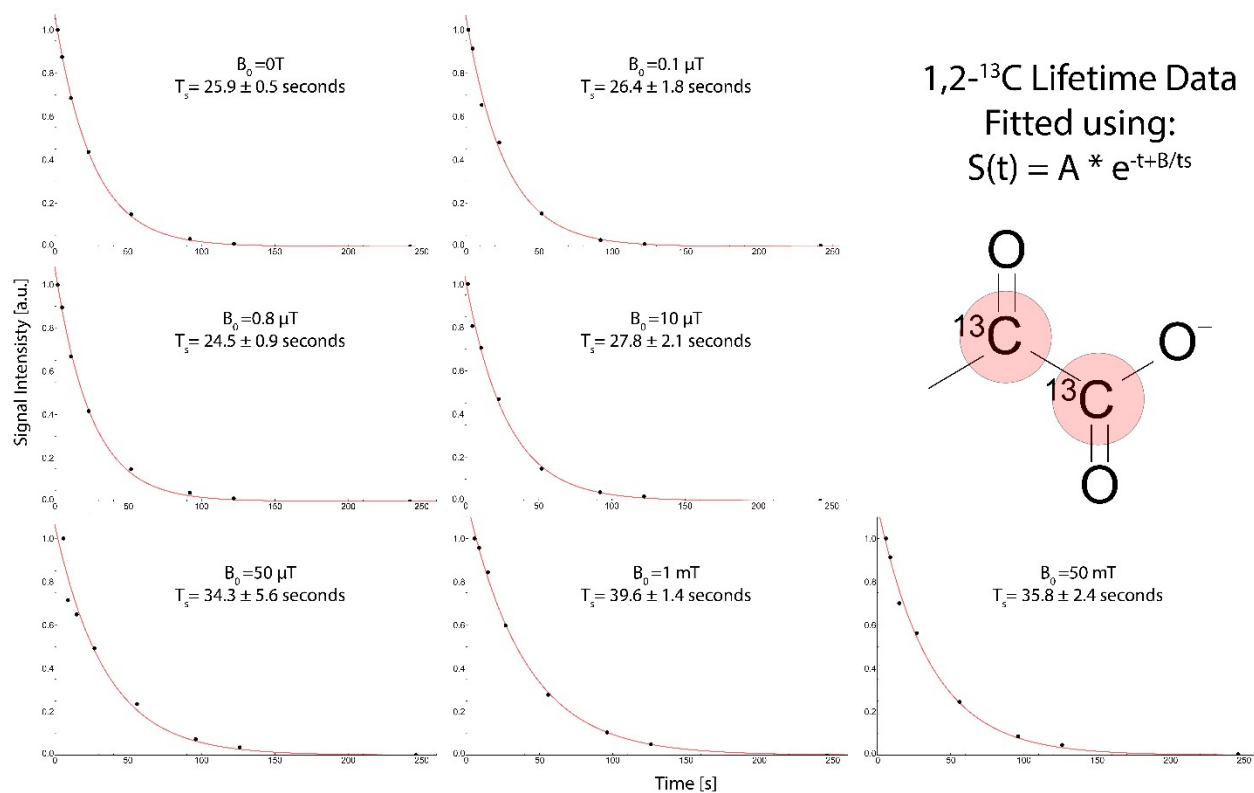


Figure S3. Lifetime data for the intensity difference on carbon-1 and carbon-2 for $[1,2-^{13}\text{C}_2]$ pyruvate at varied magnetic fields, showing a maximum T_s of 39.6 s at 1 mT. Data is only shown up to 50 mT where the singlet state remains an eigenstate of the spin system.

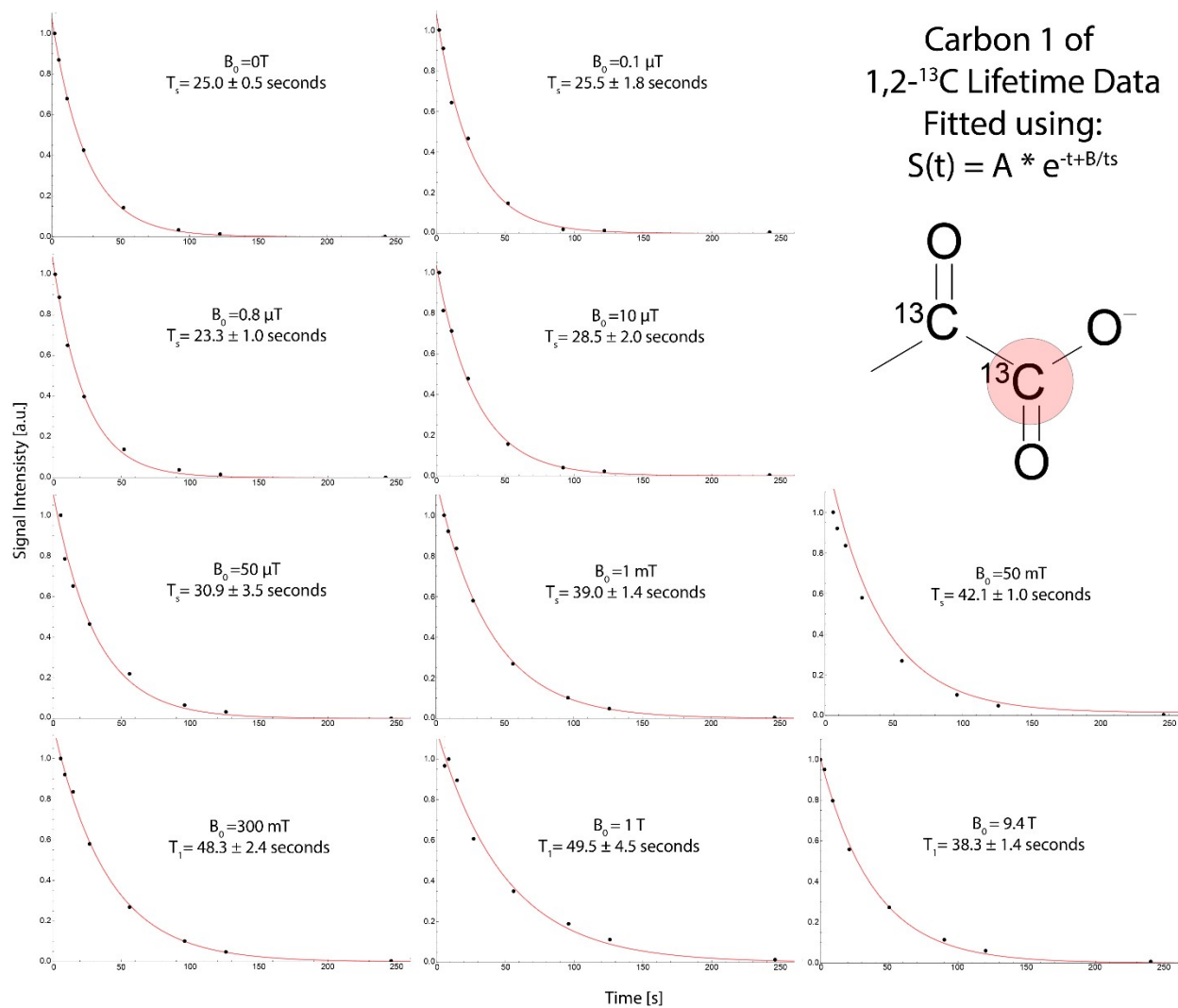


Figure S4. Lifetime data for carbon 1 of [1,2-¹³C₂]pyruvate at varied magnetic fields, showing a maximum T_1 of 49.5 s at 1 T. At magnetic fields of 50 mT and below, the measurements reflect T_s because the spins are entangled, whereas at magnetic fields of 300 mT and above, the spins behave as individual spin-1/2 particles and T_1 is observed, hence the change in label in the panels.

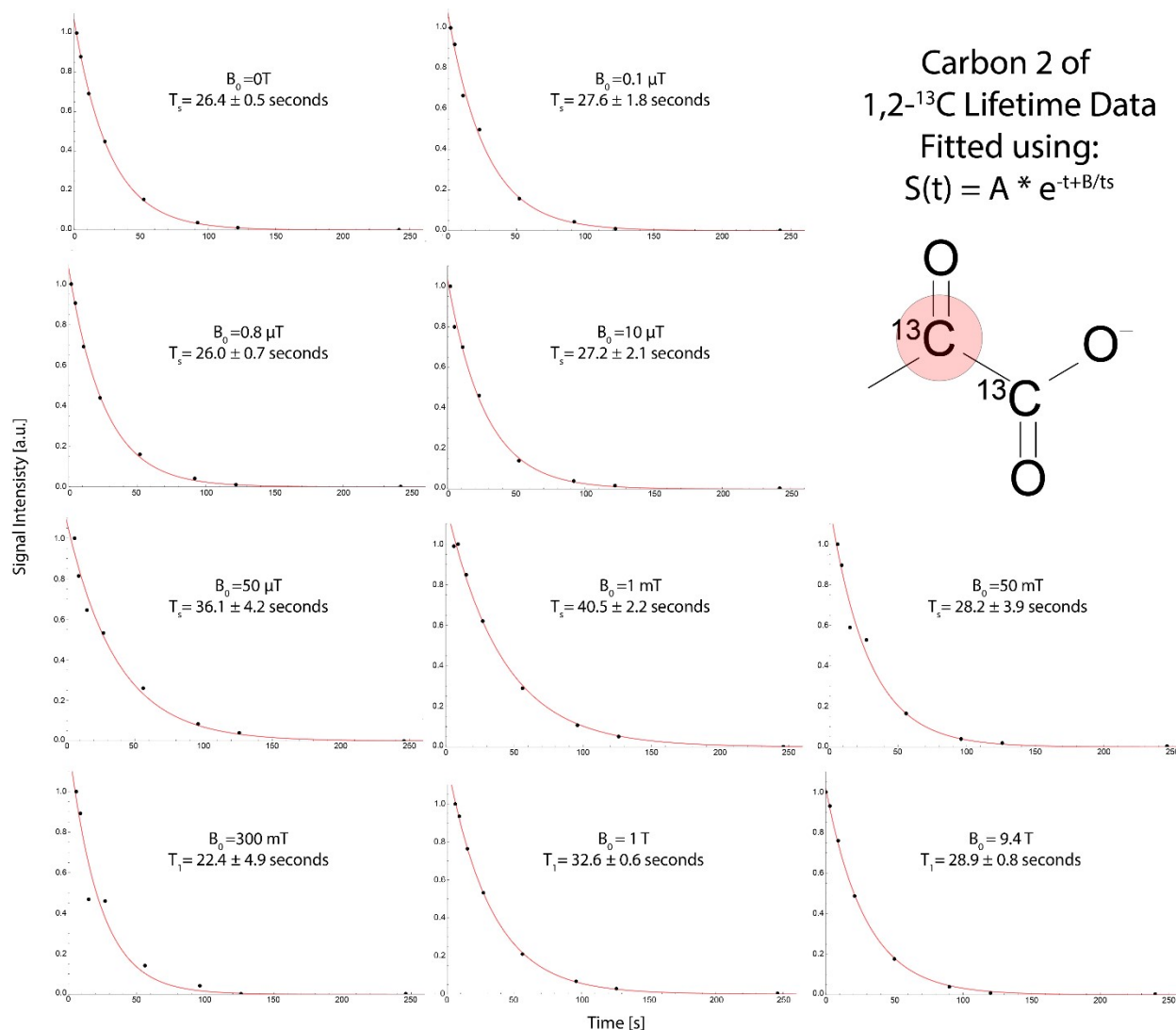


Figure S5. Lifetime data for carbon-2 in $[1,2-^{13}\text{C}_2]$ pyruvate at varied magnetic fields, showing a maximum of $T_S = 40.5$ s at 1 mT. At magnetic fields of 50 mT and below, the measurements reflect T_S because the spins are entangled, whereas at magnetic fields of 300 mT and above, the spins behave as individual spin-1/2 particles and T_1 is observed, hence the change in label in the panels.

4) 4 Spin Matrix of $[1,2-^{13}\text{C}_2]$ Pyruvate

In the paper portions of the 4 spin matrix of $[1,2-^{13}\text{C}_2]$ pyruvate was shown where the relevant terms were studied. Presented here is the complete matrix with all interactions accounted for with the on diagonal terms highlighted by a green line. Here the on and off diagonal terms can be evaluated to determine any spin state transition from the hydrides to the pyruvate system.

$ S_0^H S_0^C\rangle$	$-(J_{HS} + J_{CS})$	$\frac{\Delta J_{HS} - \Delta J_{CS}}{4}$	$\frac{\Delta J_{HS} - \Delta J_{CS}}{4}$	$\frac{\Delta J_{CS} - \Delta J_{HS}}{4}$	0	0	0	0
$ T_0^H T_0^C\rangle$	$\frac{\Delta J_{HS} - \Delta J_{CS}}{4}$	0	$\frac{\Sigma J_{HS} + \Sigma J_{CS}}{4}$	$\frac{\Sigma J_{HS} + \Sigma J_{CS}}{4}$	0	0	0	0
$ T_+^H T_-^C\rangle$	$\frac{\Delta J_{HS} - \Delta J_{CS}}{4}$	$\frac{\Sigma J_{HS} + \Sigma J_{CS}}{4}$	$(v_{11} - v_{12}) - \frac{\Sigma J_{HS} + \Sigma J_{CS}}{4}$	0	0	0	0	0
$ T_-^H T_+^C\rangle$	$\frac{\Delta J_{HS} - \Delta J_{CS}}{4}$	$\frac{\Sigma J_{HS} + \Sigma J_{CS}}{4}$	0	$(v_{11} - v_{12}) + \frac{\Sigma J_{HS} + \Sigma J_{CS}}{4}$	0	0	0	0
$ T_+^H T_+^C\rangle$	0	0	0	0	$\frac{\Sigma J_{HS} + \Sigma J_{CS}}{4} + v_{11} + v_{12}$	0	0	0
$ T_-^H T_-^C\rangle$	0	0	0	0	0	$\frac{\Sigma J_{HS} + \Sigma J_{CS}}{4} - (v_{11} + v_{12})$	0	0
$ T_0^H T_+^C\rangle$	0	0	0	0	0	0	v_{11}	$\frac{\Sigma J_{HS} - \Sigma J_{CS}}{4}$
$ T_+^H T_0^C\rangle$	0	0	0	0	0	0	$\frac{\Sigma J_{HS} + \Sigma J_{CS}}{4}$	v_{12}
$ T_0^H T_-^C\rangle$	0	0	0	0	0	0	0	0
$ T_+^H T_-^C\rangle$	0	0	0	0	0	0	0	0
$ S_0^H T_0^C\rangle$	0	0	$\frac{\Delta J_{HS} + \Delta J_{CS}}{4}$	$-\frac{\Delta J_{HS} - \Delta J_{CS}}{4}$	0	0	0	0
$ T_0^H S_0^C\rangle$	0	0	$\frac{\Sigma J_{HS} - \Sigma J_{CS}}{4}$	$\frac{\Sigma J_{HS} - \Sigma J_{CS}}{4}$	0	0	0	0
$ S_0^H T_+^C\rangle$	0	0	0	0	0	0	$-\frac{\Delta J_{HS} - \Delta J_{CS}}{4}$	$\frac{\Delta J_{HS} + \Delta J_{CS}}{4}$
$ T_+^H S_0^C\rangle$	0	0	0	0	0	0	$\frac{\Sigma J_{HS} - \Sigma J_{CS}}{4}$	$\frac{\Sigma J_{HS} - \Sigma J_{CS}}{4}$
$ S_0^H T_-^C\rangle$	0	0	0	0	0	0	0	0
$ T_-^H S_0^C\rangle$	0	0	0	0	0	0	0	0
$ T_0^H T_0^C\rangle$	0	0	0	0	0	0	0	0
$ T_0^H T_+^C\rangle$	0	0	0	0	0	0	0	0
$ T_+^H T_0^C\rangle$	0	0	0	0	0	0	0	0
$ T_+^H T_+^C\rangle$	0	0	0	0	0	0	0	0
$ T_+^H T_-^C\rangle$	0	0	0	0	0	0	0	0
$ T_-^H T_+^C\rangle$	0	0	0	0	0	0	0	0
$ T_-^H T_-^C\rangle$	0	0	0	0	0	0	0	0
$ S_0^H S_0^C\rangle$	0	0	0	0	0	0	0	0
$ T_0^H S_0^C\rangle$	0	0	0	0	0	0	0	0
$ S_0^H T_+^C\rangle$	0	0	0	0	0	0	0	0
$ T_+^H S_0^C\rangle$	0	0	0	0	0	0	0	0
$ S_0^H T_-^C\rangle$	0	0	0	0	0	0	0	0
$ T_-^H S_0^C\rangle$	0	0	0	0	0	0	0	0
$ T_0^H T_0^C\rangle$	0	0	0	0	0	0	0	0
$ T_0^H T_+^C\rangle$	0	0	0	0	0	0	0	0
$ T_+^H T_0^C\rangle$	0	0	$\frac{\Delta J_{HS} + \Delta J_{CS}}{4}$	$\frac{\Sigma J_{HS} - \Sigma J_{CS}}{4}$	0	0	0	0
$ T_-^H T_0^C\rangle$	0	0	$-\frac{\Delta J_{HS} - \Delta J_{CS}}{4}$	$\frac{\Sigma J_{HS} - \Sigma J_{CS}}{4}$	0	0	0	0
$ T_+^H T_+^C\rangle$	0	0	0	0	0	0	0	0
$ T_-^H T_+^C\rangle$	0	0	0	0	0	0	0	0
$ T_+^H T_-^C\rangle$	0	0	0	0	0	0	0	0
$ T_-^H T_-^C\rangle$	0	0	0	0	0	0	0	0
$ T_0^H T_+^C\rangle$	0	0	0	0	$-\frac{\Delta J_{HS} - \Delta J_{CS}}{4}$	$\frac{\Sigma J_{HS} - \Sigma J_{CS}}{4}$	0	0
$ T_+^H T_0^C\rangle$	0	0	0	0	$\frac{\Delta J_{HS} + \Delta J_{CS}}{4}$	$\frac{\Sigma J_{HS} - \Sigma J_{CS}}{4}$	0	0
$ T_0^H T_-^C\rangle$	$-v_{11}$	$\frac{\Sigma J_{HS} + \Sigma J_{CS}}{4}$	0	0	0	0	$\frac{\Delta J_{HS} + \Delta J_{CS}}{4}$	$\frac{\Sigma J_{HS} - \Sigma J_{CS}}{4}$
$ T_-^H T_0^C\rangle$	$\frac{\Sigma J_{HS} + \Sigma J_{CS}}{4}$	$-v_{12}$	0	0	0	0	$-\frac{\Delta J_{HS} - \Delta J_{CS}}{4}$	$\frac{\Sigma J_{HS} - \Sigma J_{CS}}{4}$
$ S_0^H T_0^C\rangle$	0	0	$-J_{HS}$	$\frac{\Delta J_{HS} - \Delta J_{CS}}{4}$	0	0	0	0
$ T_0^H S_0^C\rangle$	0	0	$\frac{\Delta J_{HS} - \Delta J_{CS}}{4}$	$-J_{CS}$	0	0	0	0
$ S_0^H T_+^C\rangle$	0	0	0	0	$-J_{HS} + v_{11}$	$\frac{\Delta J_{HS} - \Delta J_{CS}}{4}$	0	0
$ T_+^H S_0^C\rangle$	0	0	0	0	$\frac{\Delta J_{HS} - \Delta J_{CS}}{4}$	$-J_{CS} + v_{12}$	0	0
$ S_0^H T_-^C\rangle$	$\frac{\Delta J_{HS} - \Delta J_{CS}}{4}$	$-\frac{\Delta J_{HS} - \Delta J_{CS}}{4}$	0	0	0	0	$-v_{11} - J_{HS}$	$\frac{\Delta J_{HS} - \Delta J_{CS}}{4}$
$ T_-^H S_0^C\rangle$	$\frac{\Sigma J_{HS} - \Sigma J_{CS}}{4}$	$\frac{\Sigma J_{HS} - \Sigma J_{CS}}{4}$	0	0	0	0	$\frac{\Delta J_{HS} - \Delta J_{CS}}{4}$	$-v_{12} - J_{CS}$

Figure S6. The full 4 Spin Matrix of [1,2-¹³C₂]Pyruvate is shown with the on diagonal terms highlighted by the green line.

5) MATLAB Simulation

The code used to produce figure 6 in the main document is presented here in its entirety. This code goes through all the necessary steps to obtain the data presented in the paper. This code can be adapted to any spin state within the confounds of the SPINACH package.

MATLAB CODE:

```
function sabre_13Cpyruvate_LFCompleteShuttlingforSI()
%Maintenance
```

```

close all

% Spin system
sys.isotopes={'1H','1H','1H','13C','13C','1H','1H'};

% Chemical shifts
inter.zeeman.scalar={2.19 2.19 2.19 203.02 169.91 -26.59 -28.56};

% Couplings inside pyruvate
inter.coupling.scalar={ 0.0 0.0 0.0 -6.25 0.95 0.0 0.0
0.0 0.0 0.0 -6.25 0.95 0.0 0.0
0.0 0.0 0.0 -6.25 0.95 0.0 0.0
0.0 0.0 0.0 0.0 59.8 -0.97 0.8
0.0 0.0 0.0 0.0 0.0 -0.5 -0.06
0.0 0.0 0.0 0.0 0.0 0.0 -10.6
0.0 0.0 0.0 0.0 0.0 0.0 0.0};

% Magnetic fields
nmr_field=9.4;

% Basis set
bas.formalism='sphten-liouv';
bas.approximation='none';

% Define the stepsizes and number of steps %this is the bubbling time
% stepsize x evolve
stepsize_bound=3.0; % stepsize of bound substrate in ms
stepsize_free=1; % stepsize of free substrate in ms
evolve_bound=10000; % Evolution steps of bound substrate in stepsize (e.g. 0.1 ms)
evolve_free=10000; % Evolution steps of free substrate in stepsize (e.g. 0.1 ms)

tau=15; % lifetime of bound complex in ms
relax=25; % lifetime of spin order in s ("Relaxation") to evolve the free species. Dont
confuse with linebroadening/apodization!

%variables for Shuttling exponentially into B0
shuttling_time=1; %shuttling time in seconds *1000 0.1=1ms
shuttling_steps=1024; %number of magnetic fields while exponentially going to the detection
field

%Do entire field spweep by rerunning entire program
sweep_steps=36; %number of steps in field sweep

for m=1:sweep_steps
    polarization_field=(m*(0.10001e-6))+(-0.1e-6);

% Do the housekeeping
sys.magnet=polarization_field;
spin_system=create(sys,inter);
spin_system=basis(spin_system,bas);

% Get the Hamiltonian superoperator
H=hamiltonian(assume(spin_system,'labframe'));

% Start in a singlet state on the hydrides
rho=singlet(spin_system,6,7);

% Evolve the system coupled to the hydrides (bound complex)
rho=evolution(spin_system,H,[],rho,stepsize_bound/1000,evolve_bound,'trajectory');

% Weight the density matrices

```

```

for k=1:evolve_bound+1
rho(:,k)=rho(:,k)*exp(-stepsize_bound*k/tau);
end

% Collect the density matrices
rho=sum(rho,2);

% Decouple hydrides
[H_D,rho]=decouple(spin_system,H,rho,[6 7]);

% Evolve the free species, weight and collect
rho=evolution(spin_system,H_D,[],rho,stepsize_free/1000,evolve_free,'trajectory');
for i=1:evolve_free+1
rho(:,i)=rho(:,i)*exp(-stepsize_free*0.001*i/relax);
end

rho=sum(rho,2);

% Split the Hamiltonian superoperator
Hz=hamiltonian(assume(spin_system,'nmr','zeeman'));

Hc=hamiltonian(assume(spin_system,'nmr','couplings'));

Hz=decouple (spin_system,Hz,[],[6 7])/polarization_field;

Hc=decouple(spin_system,Hc,[],[6 7]);

% Spin Contribution Calculations
Iden=state(spin_system,{'E' , 'E' },{4,5},'exact');
LpSm=state(spin_system,{'L+' , 'L-' },{4,5},'exact');
LmSp=state(spin_system,{'L-' , 'L+' },{4,5},'exact');
LzSz=state(spin_system,{'Lz' , 'Lz' },{4,5},'exact');

S0T=- (1/2*(LpSm+LmSp)+LzSz);
T0T=+(1/2*(LpSm+LmSp))-LzSz;
TpT=+LzSz+(1/2*(LpSm));
TmT=+LzSz-(1/2*(LpSm));

Lz=state(spin_system,{'Lz' },{4},'exact');
Sz=state(spin_system,{'Lz' },{5},'exact');
TpminTm=Lz-Sz;
TpplusTm=Lz+Sz;

SoBper=100*(S0T'*rho);
ToBper=100*(T0T'*rho);
Tplusperbefore=100*(TpT'*rho);
Tminusperbefore=100*(TmT'*rho);
TpminTmBefore=100*(TpminTm'*rho);
TpplusTmBefore=100*(TpplusTm'*rho);

pertable_before(1,m)=SoBper;
pertable_before(2,m)=ToBper;
pertable_before(3,m)=Tplusperbefore;
pertable_before(4,m)=Tminusperbefore;
pertable_before(5,m)=TpminTmBefore;
pertable_before(6,m)=TpplusTmBefore;

% Back to Field Sweep
Collect=0;

```



```

%Lift the field exponentially in 1024 steps
for field=exp(linspace(log(polarization_field),log(nmr_field),shuttling_steps))

rho=evolution(spin_system,Hc+field*Hz,[],rho,shuttling_time/(shuttling_steps),1000,'final');
Collect=[Collect; field];
end

%need to transfer to main magnetic field before detection
sys.magnet=nmr_field;
spin_system=create(sys,inter);
spin_system=basis(spin_system,bas);
%*****

% Set NMR experiment parameters
parameters.offset=19000;
parameters.spins={'13C'};
parameters.sweep=6000;
parameters.npoints=10000;
parameters.zerofill=100000;
parameters.axis_units='ppm';
parameters.invert_axis=1;

% Set the detection state and detected spins
coil=state(spin_system,'L+', '13C', 'exact');

% Get the pulse operator; choose spins
Lp=operator(spin_system,{'L+'},{4})+operator(spin_system,{'L+'},{5});

% Apply pi/2 pulse on Y axis
rho=step(spin_system,(Lp-Lp')/2i,rho,pi/2);

% Detect the magnetization on free substrate
H=hamiltonian(assume(spin_system,'nmr'));
[H_D,rho]=decouple(spin_system,H,rho,[6 7]);
H=H_D;
fid=evolution(spin_system,H,coil,rho,1/parameters.sweep,...
parameters.npoints-1, 'observable');

% Apodization: lb=k*sw/npoints
lb=3;
k_lb=parameters.npoints*lb/parameters.sweep;
fid=apodization(fid,'exp-1d',k_lb);

% Fourier transform
spectrum=fftshift(fft(fid,parameters.zerofill));

% Plotting
plotted=figure();
plot_1d(spin_system,-real(spectrum),parameters);
name=['x' num2str((m)) 'nT'];
saveas(plotted,name,'fig')
end
clear;
close all
for j = 1 : 36
a=num2str((j));
opencurrentfigure=['x' a 'nT.fig'];
h(j) = open(opencurrentfigure);
end
handleLine = findobj(h,'type','line');

```

```

hold on ;
combineplots=[get(handleLine(1),'YData')];
for i = 2 : length(handleLine)
    plot(get(handleLine(i),'XData'), get(handleLine(i),'YData'));
    combineplots = [combineplots; get(handleLine(i),'YData')];
end

hold off;
figure();
w =
waterfall(get(handleLine(i),'XData'), linspace(1,length(handleLine),length(handleLine)),combine
plots);
w.FaceAlpha = 0;
xlim([170 215])
ax = gca;
ax.XDir = 'reverse';
view(382,20)
name=['x'];
saveas(w,name,'fig')
saveas(w,name,'png')

close all

```

6) Individual Spin State Contribution

Here we show the individual spin state contributions of $S_0 - T_0$ and $T_+ + T_-$ as a function of magnetic field. These values were found through simulations and the code is presented above in section 5 of the SI under the subheading “% Spin Contribution Calculations”. As discussed in the main text the contributions of these spin states combine to produce the simulated waterfall plot shown, in Figure 7B., in the main text.

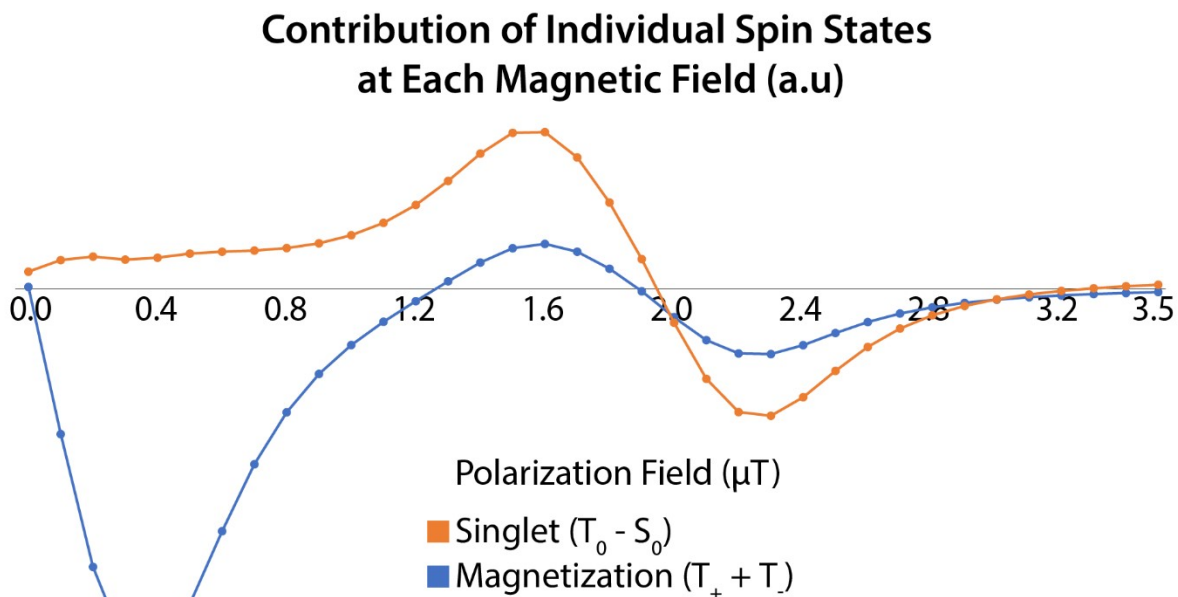


Figure S7. Individual spin state contributions of $S_0 - T_0$ and $T_+ + T_-$ shown as intensity (a.u.) [S7].