**Supporting Information**

# Enhancement of Weak Signals by Suppressing High-Intense Methyl and Methylene Signals of Lipids in NMR Spectroscopy

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## NMR acquisition parameters for measurements

**Table S1. Experimental data for cheese, butter, a stock solution of lipids, coconut oil, and olive oil samples.**

| Experiment Name | Acq (s) | $D_1$ (s) | SW (ppm) | Data points | NS | FID Res. (Hz) | Zero Filling | LB Hz | Δ ms | WDW | Expt Time |
|---|---|---|---|---|---|---|---|---|---|---|---|
| 1D $^1$H | 2.0 | 10 | 20.15 | 64516 | 64 | 0.5 | 131K | 0.3 | - | EM | 13 m, 38 s |
| 1D $^1$H-SMLS | | | | | - | - | - | - | 20 | - | 13 m, 51 s |
| TOCSY | 0.299 | 1.5 | 12.01 | 5768 | 8 | 3.33 | 8192 | 1.0 | - | QSINE | 2 h, 33 m, 40 s |
| SMLS-TOCSY | 0.031 | - | 12.01 | 600 | - | 32.05 | 1024 | 0.3 | 20 | QSINE | 2 h, 37 m, 51 s |
| TOCSY-SUN | 0.299 | 1.5 | 12.01 | 5768 | 8 | 3.33 | 8192 | 1.0 | - | QSINE | 2 h, 44 m, 31 s |
| | 0.031 | - | 12.01 | 600 | - | 32.05 | 1024 | 0.3 | | QSINE | |

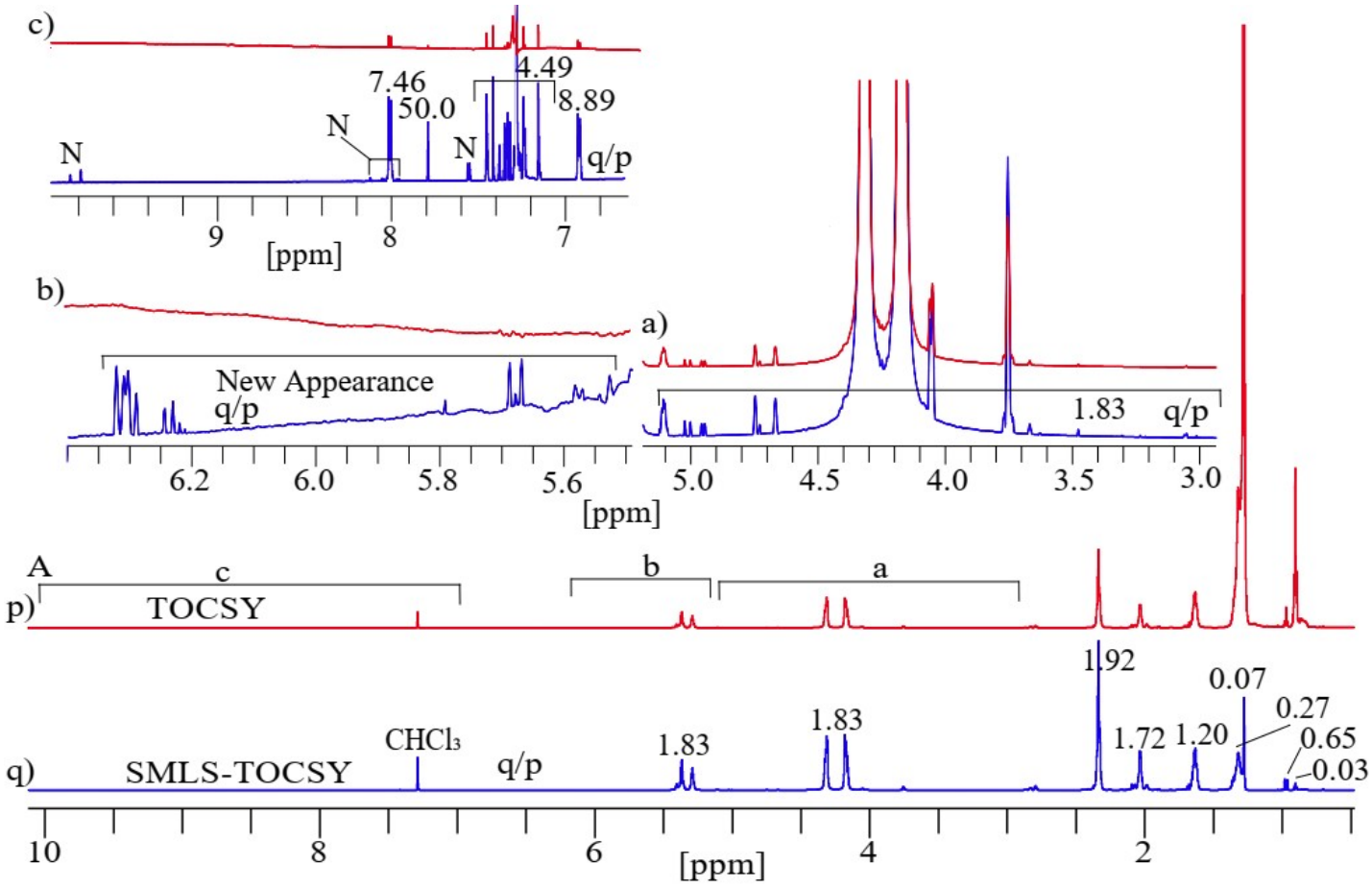
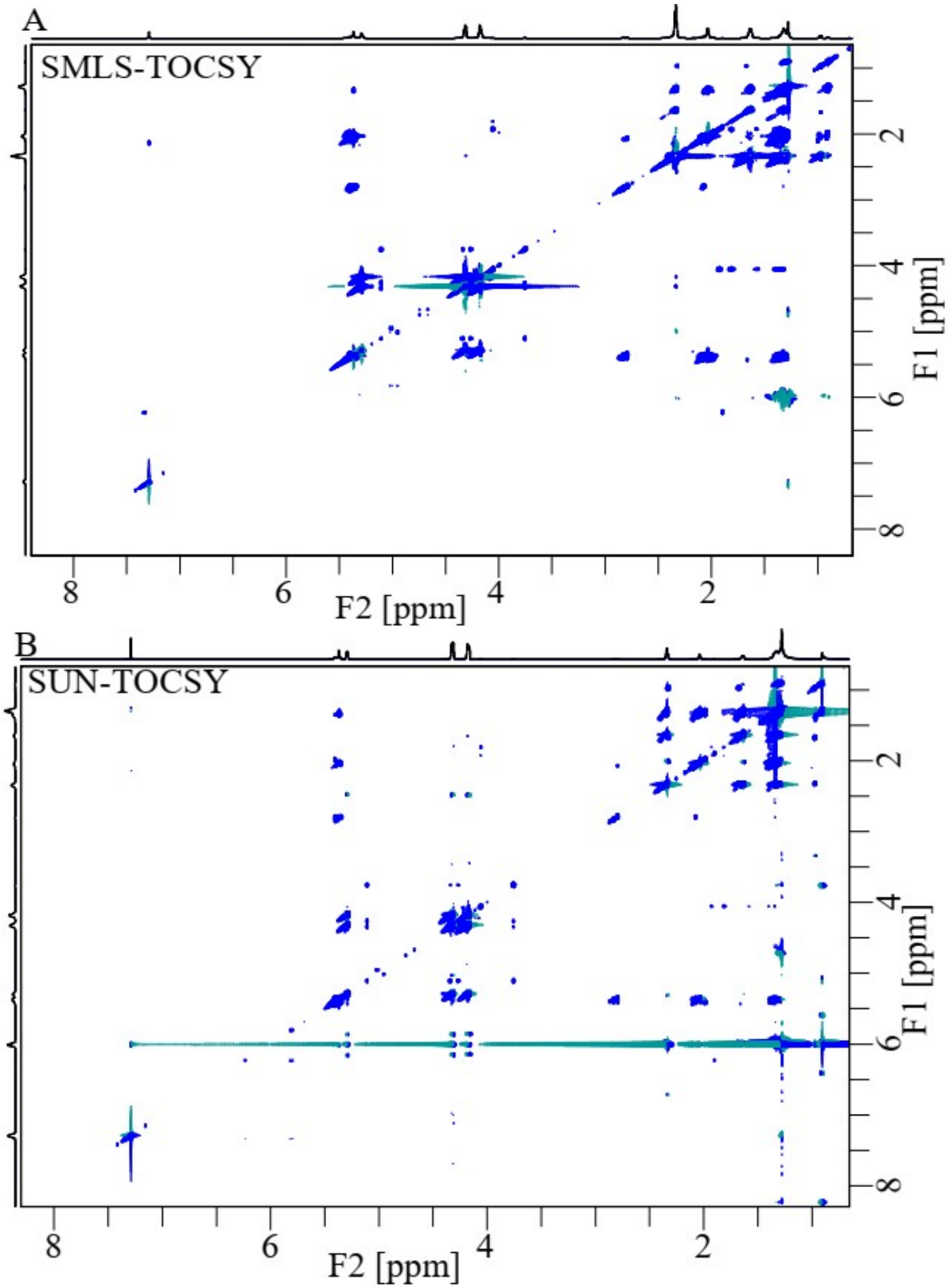



Fig. S1. (A) Typical projected 1D $^{1}$H of TOCSY (p), and SMLS-TOCSY (q) of cheese sample divided into the sections of a, b, and c those were shown in expanded spectra of regions 3.00 - 5.10 ppm (a), 5.50 - 6.40 ppm (b), and 6.78 – 9.98 ppm (c) with estimated peak intensities in numbers and N is for new.




**Comparison between 2D SLMS-TOCSY and TOCSY-SUN in Cheese Sample**

Fig. S2. represents the 2D NMR spectra of SMLS-TOCSY in (A), and TOCSY-SUN in (B) of cheese sample.

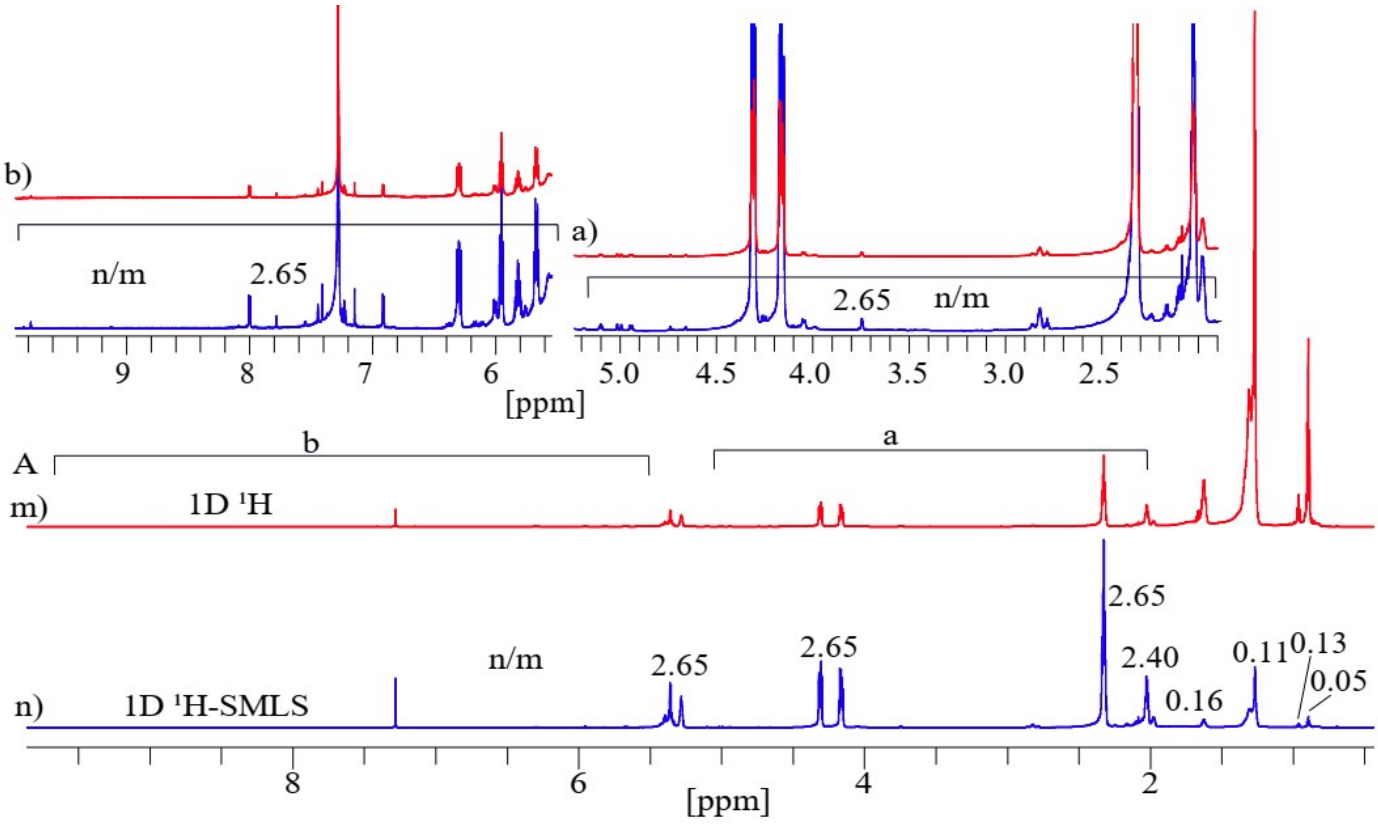



Fig. S3. (A) Typical 1D $^{1}$H (m), and $^{1}$H-SMLS (n) of butter sample divided into the sections of a, and b those were shown in expanded spectra of regions 1.95 - 5.20 ppm (a), and 5.60 - 9.90 ppm (b) with estimated peak intensities in numbers.

**Comparison between 2D SLMS-TOCSY and TOCSY-SUN in Butter Sample**

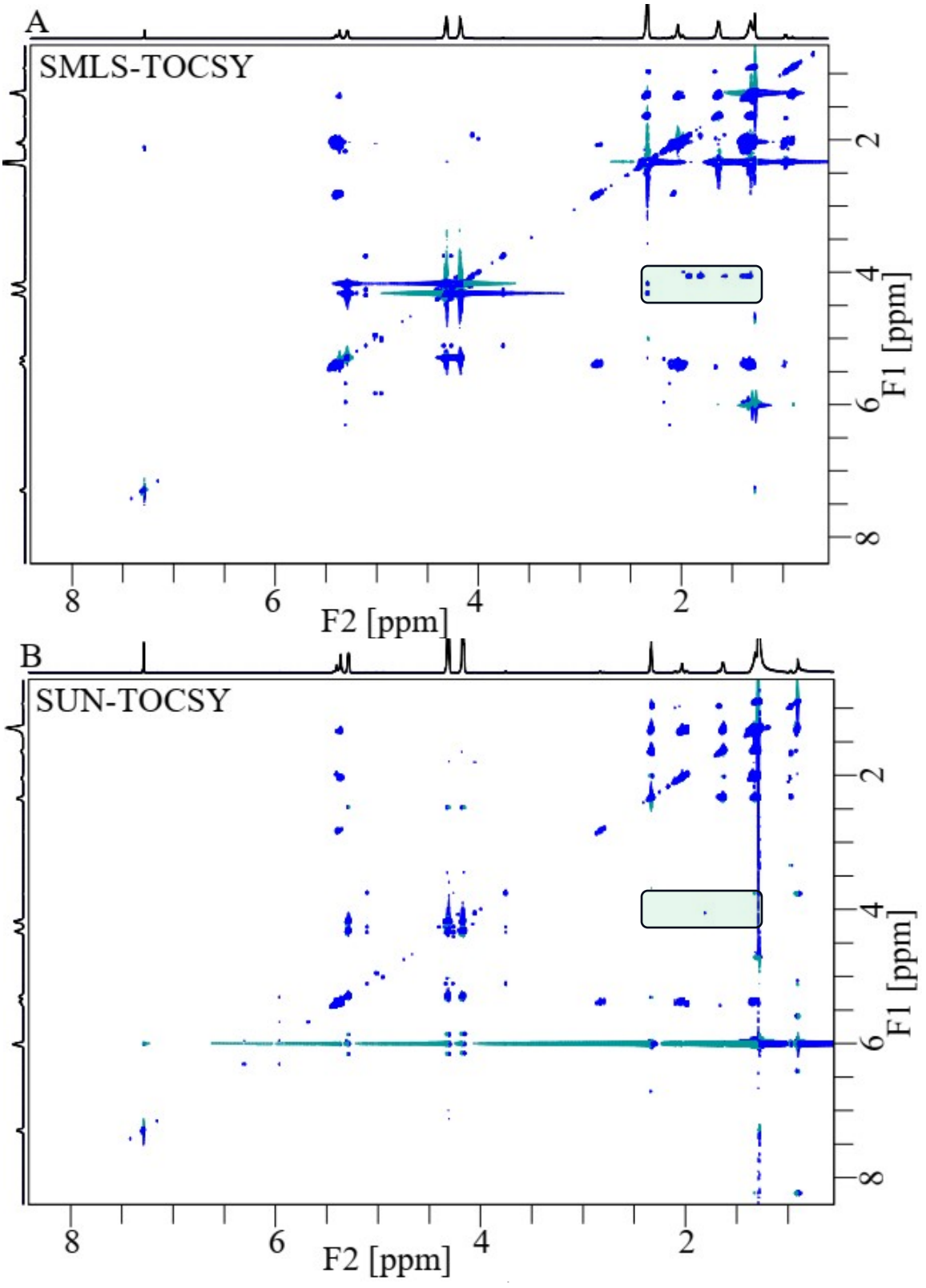


Fig. S4. represents the 2D NMR spectra of SMLS-TOCSY in (A), and TOCSY-SUN in (B) of butter sample.

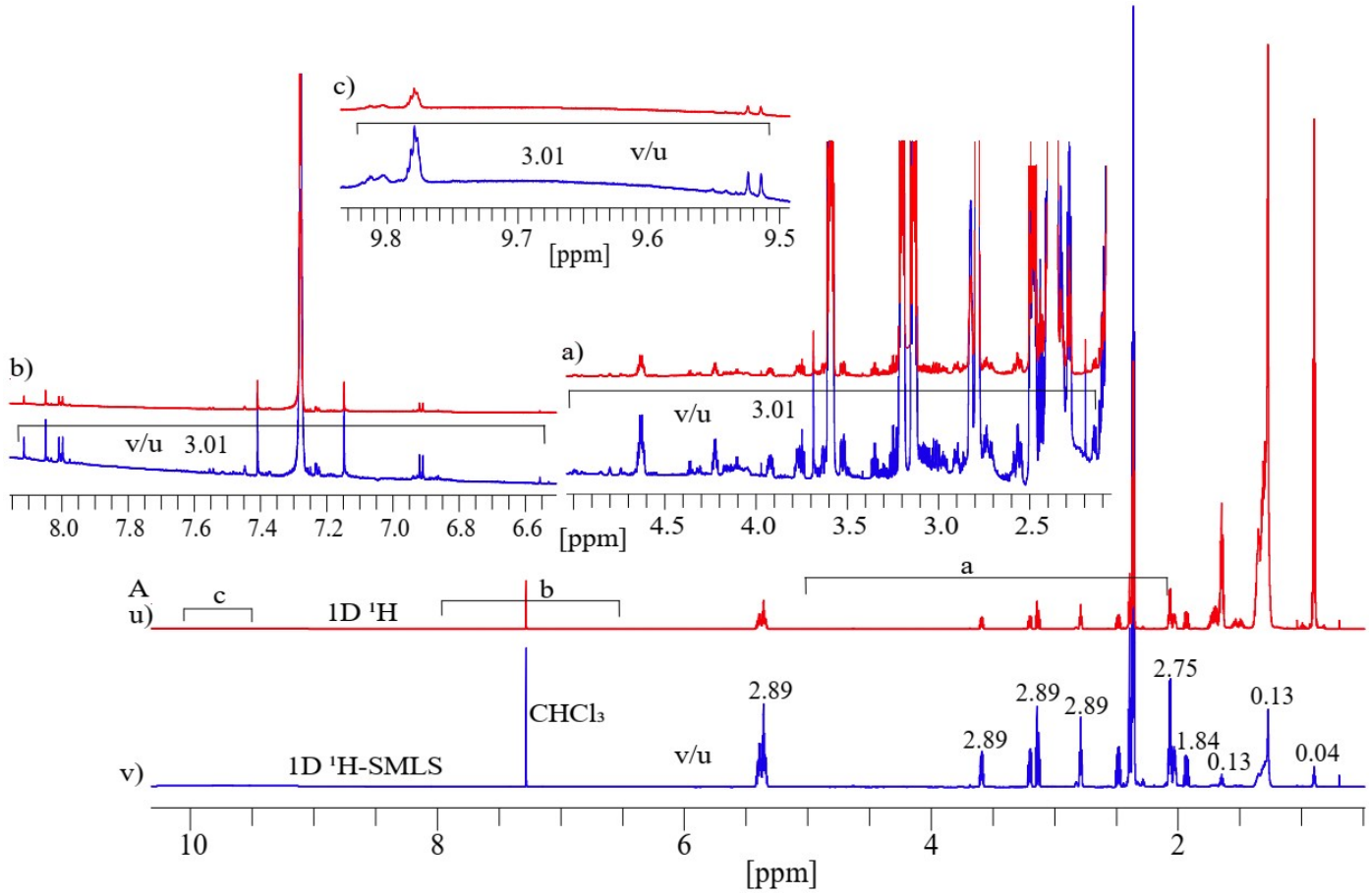
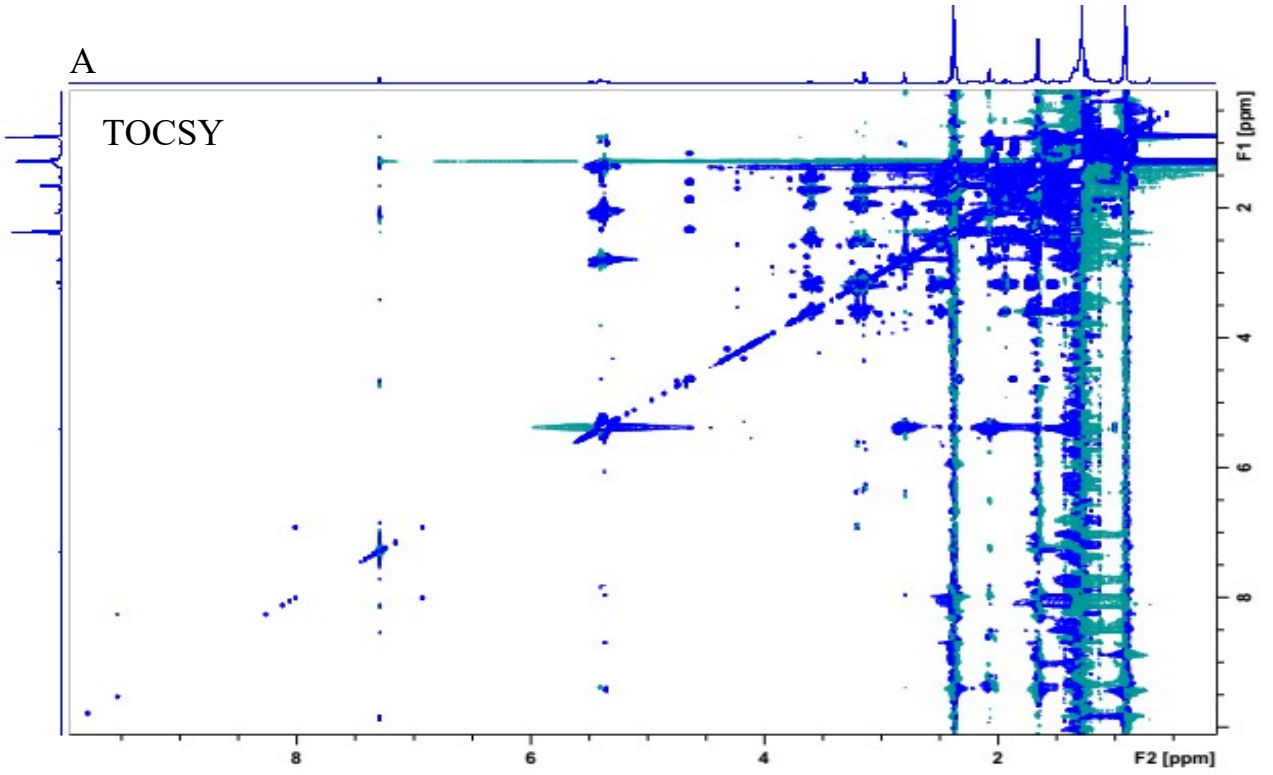



Fig. S5. (A) Typical 1D $^{1}$H (u), and $^{1}$H-SMLS (v) of a stock solution of lipids sample divided into the sections of a, b, and c those were shown in expanded spectra of regions 2.10 - 5.00 ppm (a), 6.65 - 8.95 ppm (b), and 9.50 – 9.83 ppm (c) with estimated peak intensities in numbers.


A
TOCSY
F1 [ppm]
F2 [ppm]
8
6
4
2

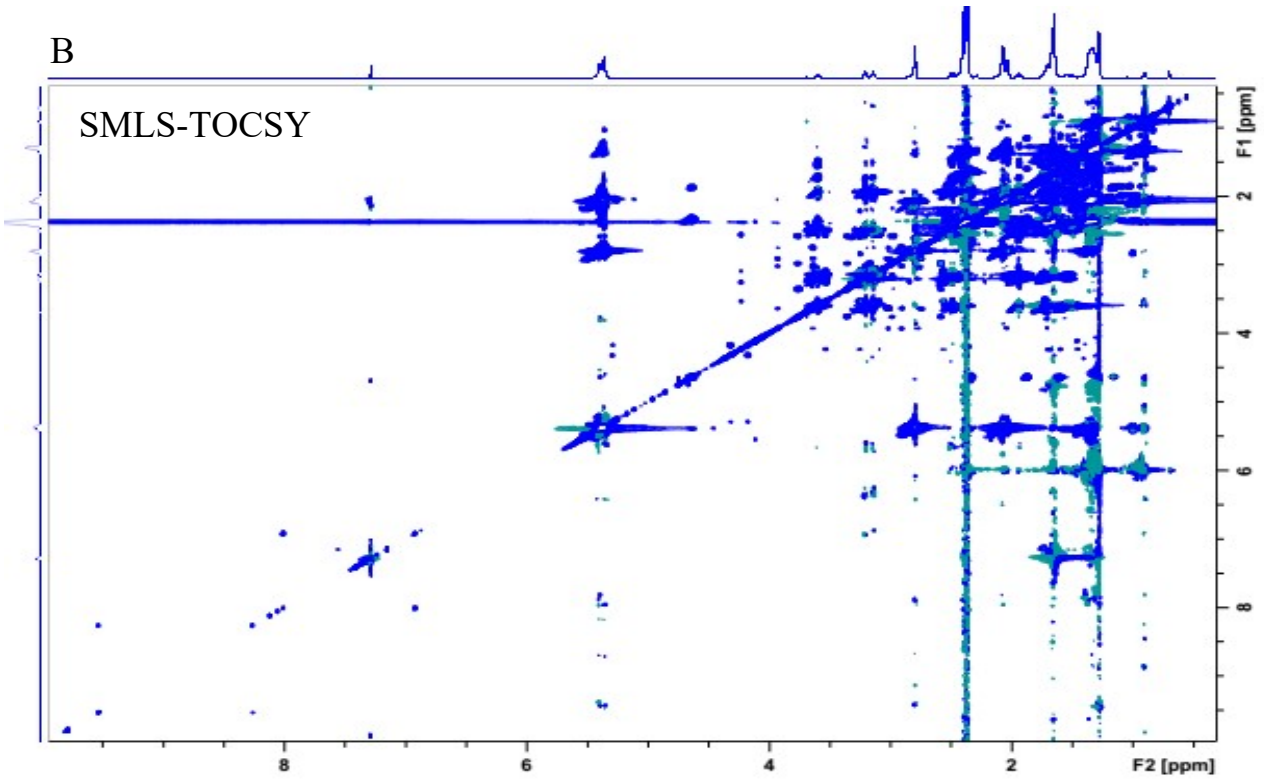

B
SMLS-TOCSY
F1 [ppm]
F2 [ppm]
8
6
4
2

Fig. S6. represents the 2D NMR spectra of TOCSY in (A), and SMLS-TOCSY in (B) of a sample prepared of a stock solution of few lipids with known concentrations in $CDCl_3$ solvent.

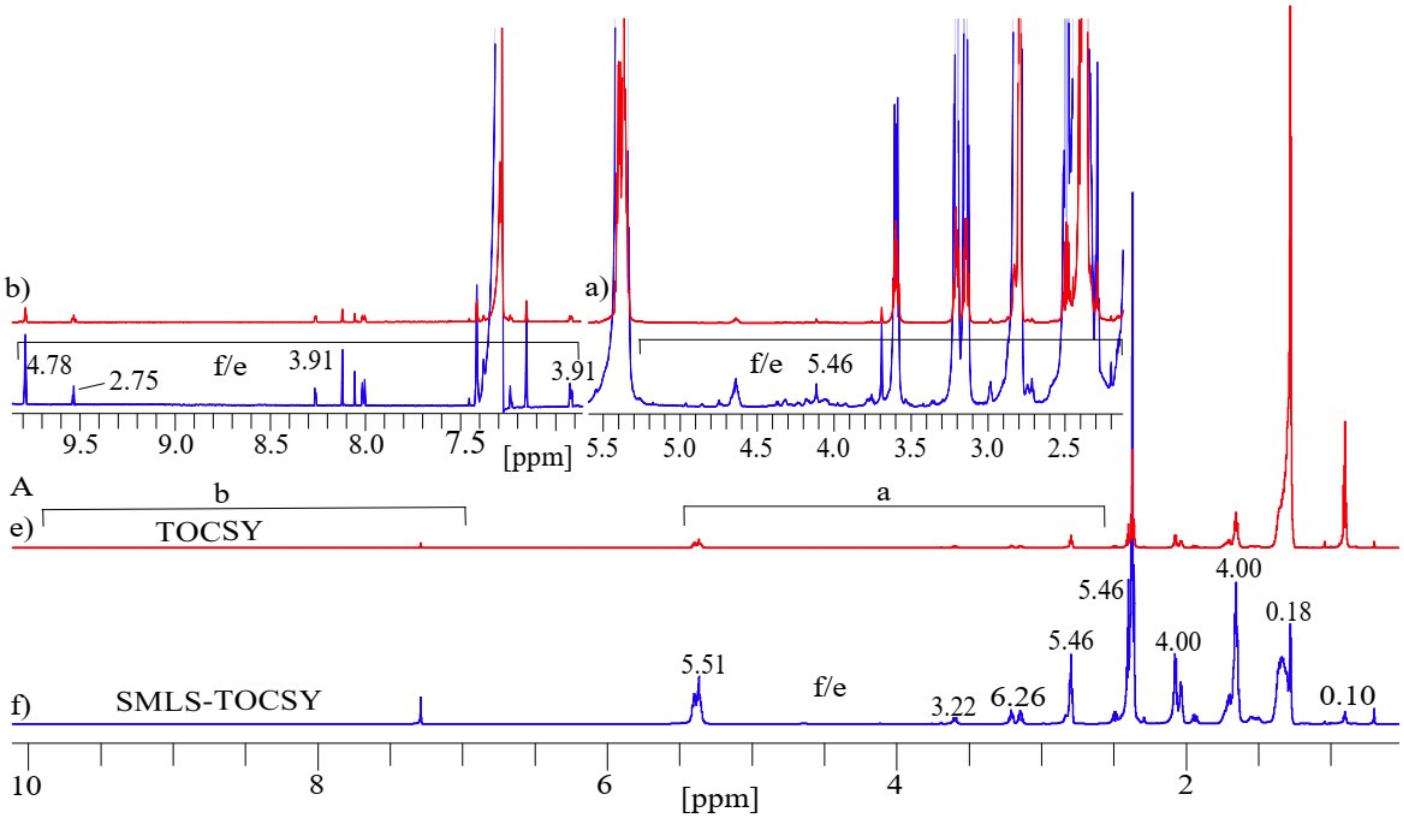



Fig. S7. (A) Typical projected 1D $^1H$ of TOCSY (e), and SMLS-TOCSY (f) of a stock solution of lipids sample divided into the sections of a, and b those were shown in expanded spectra of regions 2.18 - 5.50 ppm (a), and 7.00 - 9.81 ppm (b) with estimated peak intensities in numbers.

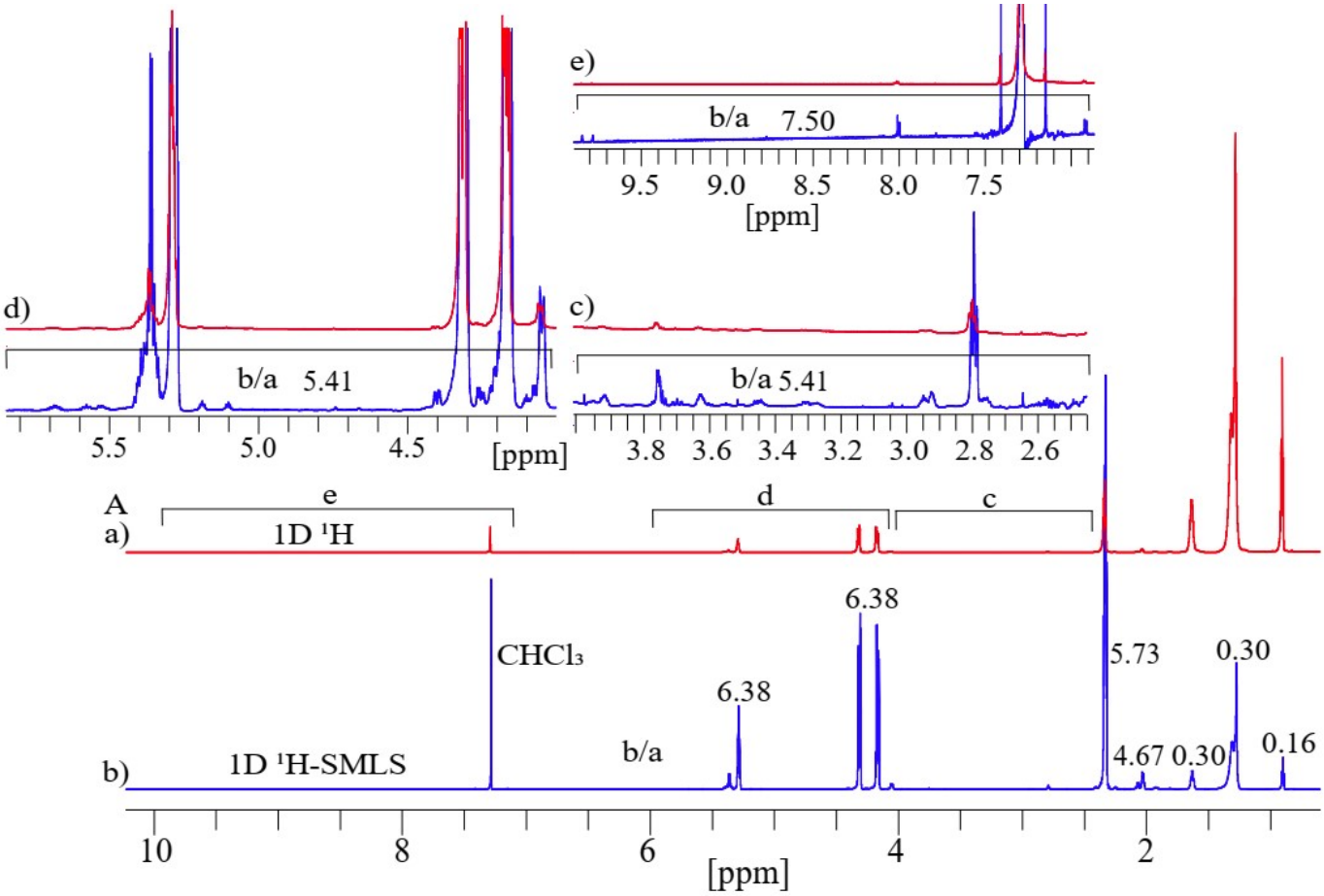



Fig. S8. (A) Typical 1D $^{1}$H (a), and $^{1}$H-SMLS (b) of coconut oil sample divided into the sections of c, d, and e were shown in expanded spectra of regions 2.45 - 4.00 ppm (c), 4.01 - 5.85 ppm (d), and 6.95 – 9.85 ppm (e) with estimated peak intensities in numbers.

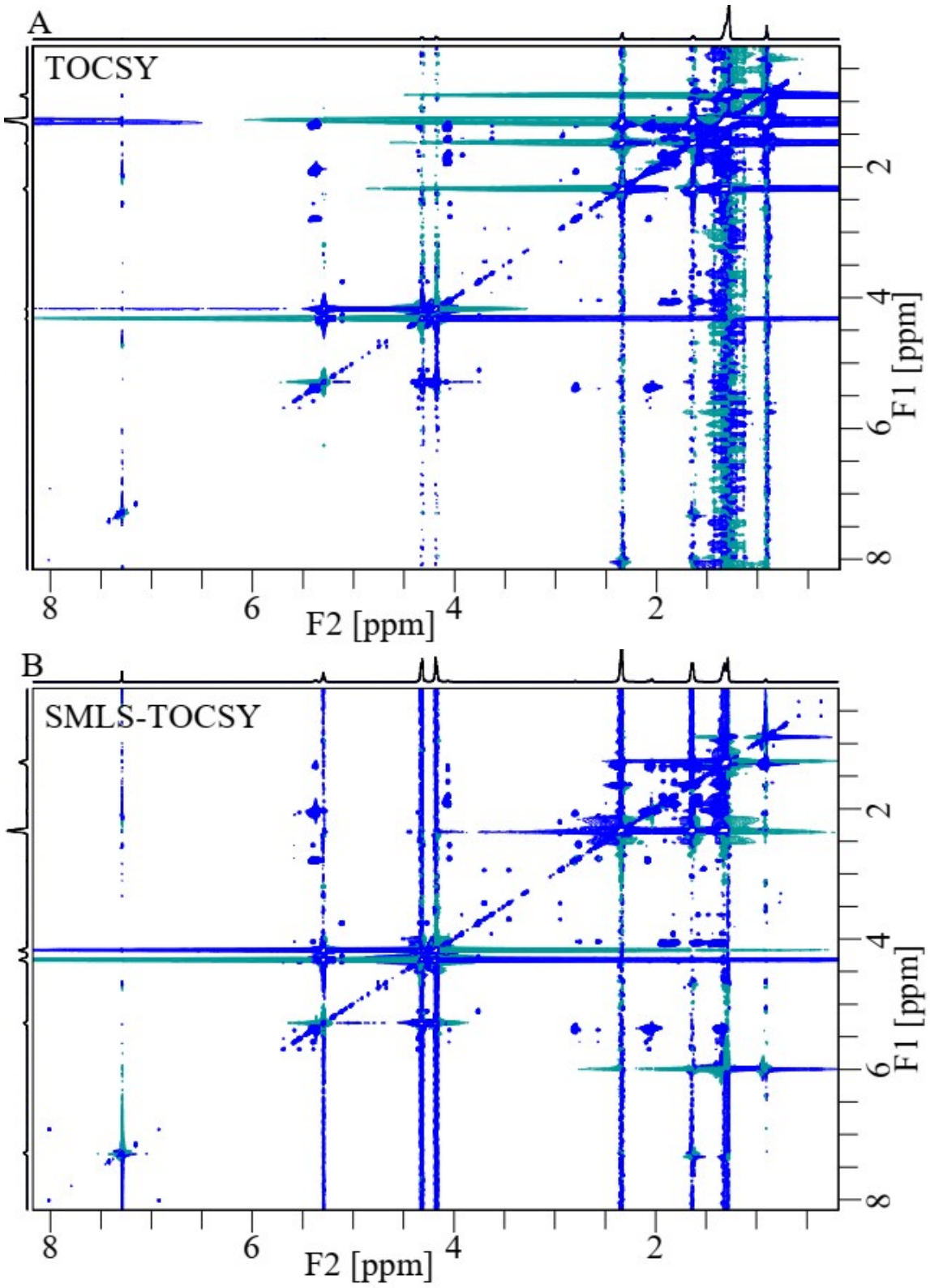



Fig. S9. displays the 2D NMR spectra of TOCSY in (A), and SMLS-TOCSY in (B) of coconut oil sample dissolved in $CDCl_3$ solvent. Suppression of methylene groups resulted in significantly enhanced visibility in (B).

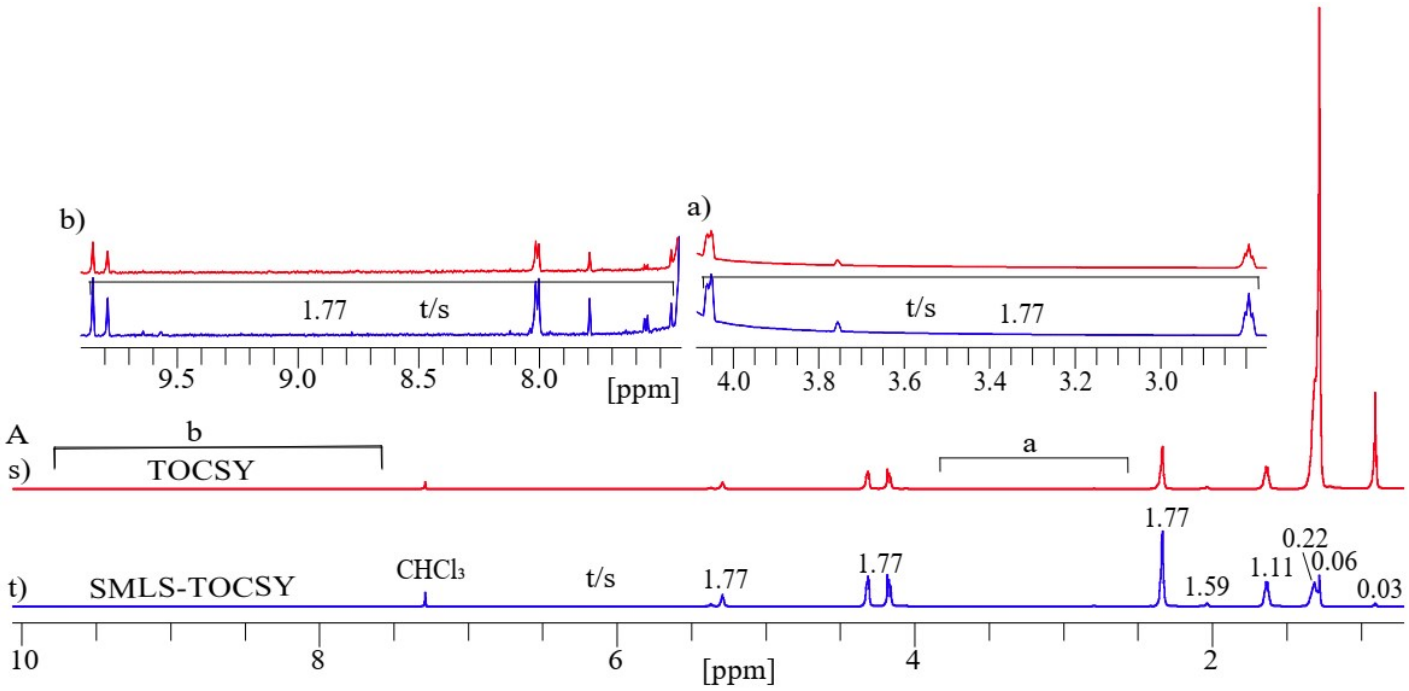



Fig. S10. (A) Typical projected 1D $^{1}$H of TOCSY (s), and SMLS-TOCSY (t) of coconut oil sample divided into the sections of a, and b those were shown in expanded spectra of regions 2.80 - 4.05 ppm (a), and 7.50 - 9.85 ppm (b) with estimated peak intensities in numbers.

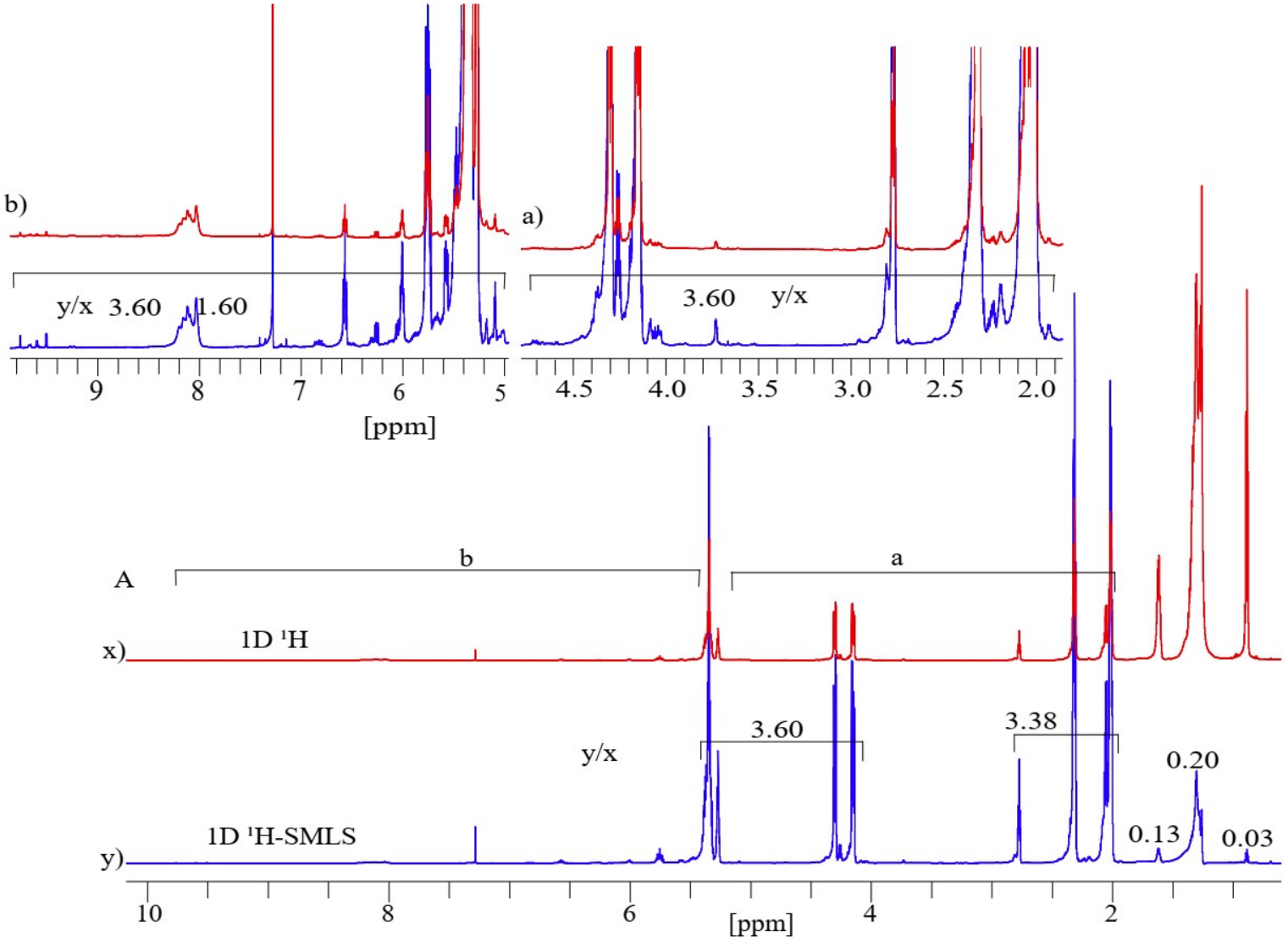



Fig. S11. (A) Typical 1D $^{1}$H (x), and $^{1}$H-SMLS (y) of olive oil sample divided into the sections of a, and b those were shown in expanded spectra of regions 1.90 - 4.70 ppm (a), and 5.00 - 9.80 ppm (b) with estimated peak intensities in numbers.

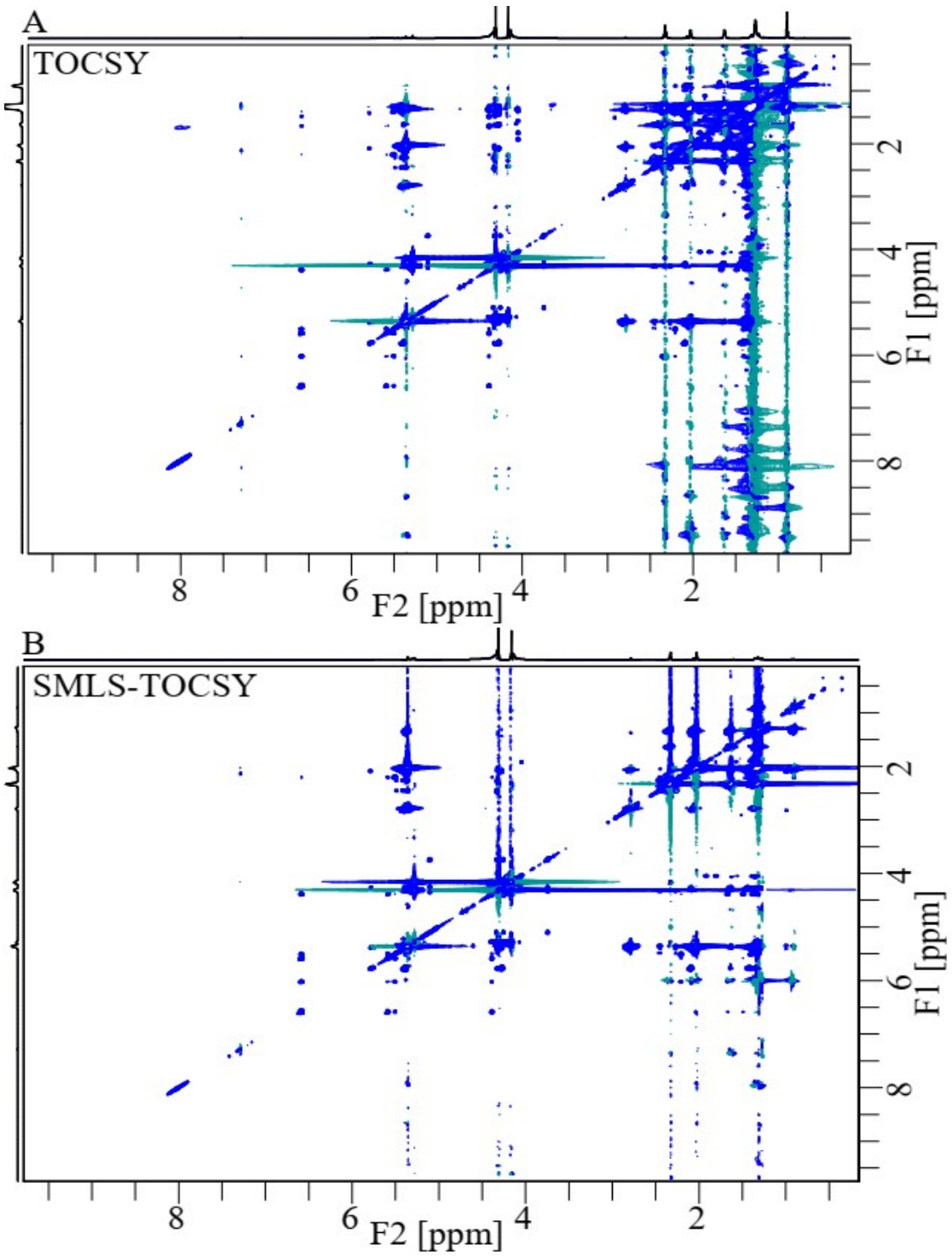



Fig. S12. represents the 2D NMR spectra of TOCSY in (A), and SMLS-TOCSY in (B) of the olive oil sample. Suppression in the conventional method is significant resulting in greatly increased and clear visibility of cross-peaks.

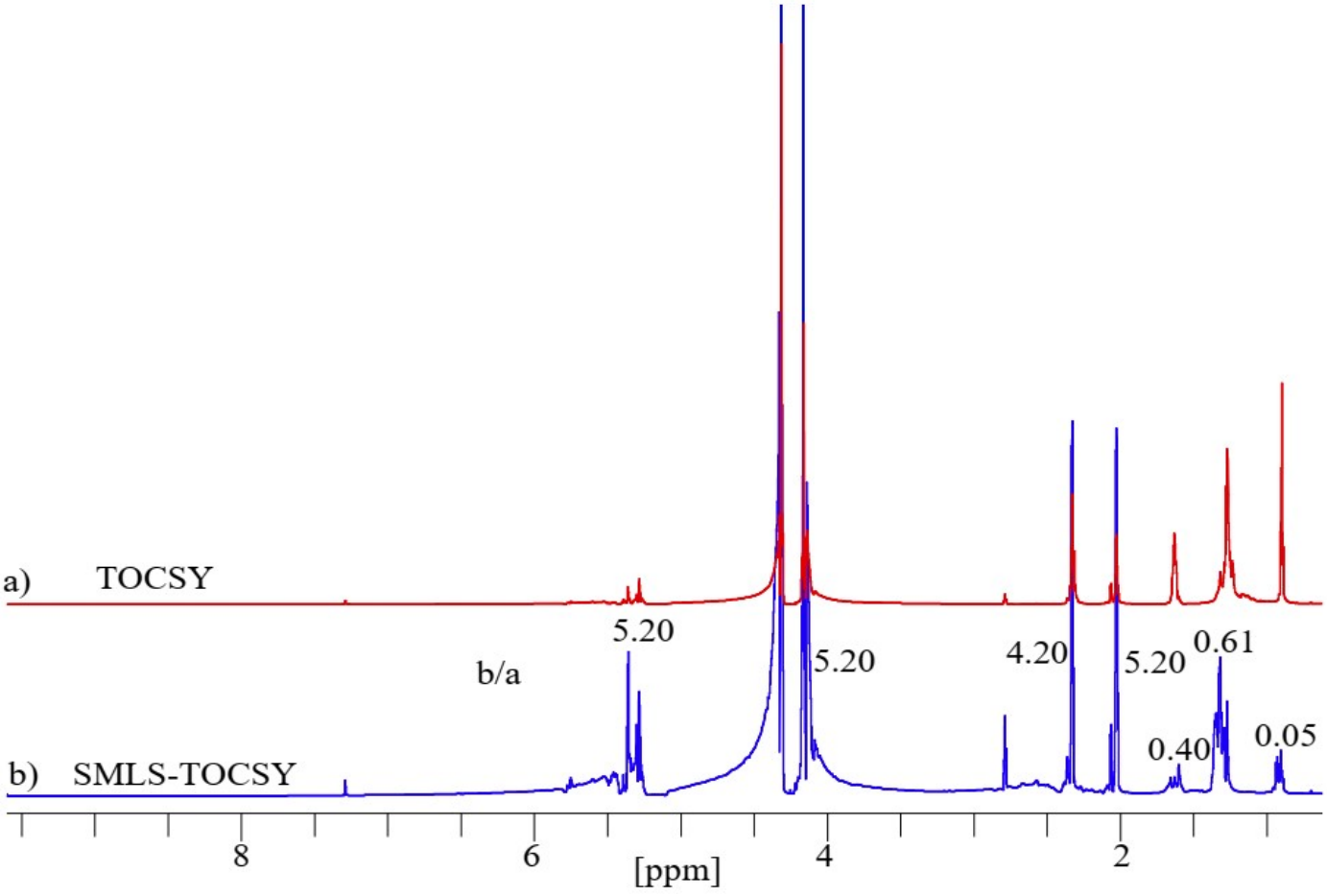



Fig. S13. (A) Typical projected 1D $^{1}$H of TOCSY (a), and SMLS-TOCSY (b) of olive oil sample with estimated peak intensities in numbers.

**Table S2. Residue intensity of suppressed peaks and Increased intensity of other peaks.**

| Types of Experiments | Types of Samples | Methyl Group (Supp.Res.Int.) | Methylene Group (Supp.Res.Int.) | Aliphatic Unsaturated groups (Increa. Int.) | Aromatic Unsaturated groups (Increa. Int.) |
|---|---|---|---|---|---|
| | Cheese | | | | |
| 1D $^{1}$H | | 1 | 1 | 1 | 1 |
| $^{1}$H-SMLS | | 0.08 | 0.20 | 2.77 | 2.77 |
| Proj 1D $^{1}$H TOCSY | | 1 | 1 | 1 | 1 |
| Proj 1D $^{1}$H SMLS-TOCSY | | 0.03 | 0.27 | 1.83 | Av 6.94 |
| | Butter | | | | |
| 1D $^{1}$H | | 1 | 1 | 1 | 1 |
| $^{1}$H-SMLS | | 0.05 | 0.11 | 2.65 | 2.65 |
| Proj 1D $^{1}$H TOCSY | | 1 | 1 | 1 | 1 |
| Proj 1D $^{1}$H SMLS-TOCSY | | 0.05 | 0.33 | Av 3.13 | Av 3.54 |
| | ASSL | | | | |
| 1D $^{1}$H | | 1 | 1 | 1 | 1 |
| $^{1}$H-SMLS | | 0.04 | 0.13 | 3.01 | 3.01 |
| Proj 1D $^{1}$H TOCSY | | 1 | 1 | 1 | 1 |
| Proj 1D $^{1}$H SMLS-TOCSY | | 0.10 | 0.18 | 5.46 | 3.91 |
| | Coco. Oil | | | | |
| 1D $^{1}$H | | 1 | 1 | 1 | 1 |
| $^{1}$H-SMLS | | 0.16 | 0.30 | 5.41 | 7.50 |
| Proj 1D $^{1}$H TOCSY | | 1 | 1 | 1 | 1 |
| Proj 1D $^{1}$H | | | | | |

| | | | | | |
|---|---|---|---|---|---|
| SMLS-TOCSY | | 0.03 | 0.22 | 1.77 | 1.77 |
| | Olive oil | | | | |
| 1D $^{1}$H | | 1 | 1 | 1 | 1 |
| $^{1}$H-SMLS | | 0.03 | 0.20 | 3.6 | 3.6 |
| Proj 1D $^{1}$H TOCSY | | 1 | 1 | 1 | 1 |
| Proj 1D $^{1}$H SMLS-TOCSY | | 0.05 | 0.61 | 5.20 | 5.20 |

**Pulse Programs of 1D $^{1}$H-SMLS, and 2D SMLS-TOCSY**

*1D $^{1}$H-presat-SMLS with band-selective suppression*

```
;1H-SMLS
;avance-version (12/01/11)
;1D sequence with f1 presaturation
;$CLASS=HighRes
;$DIM=1D
;$TYPE=
;$SUBTYPE=
;$COMMENT=
#include <Avance.incl>
#include <Grad.incl>
#include <Delay.incl>
"d12=20u"
;"TAU=50m"
"acqt0=-p1*2/3.1416"

1 ze
2 30m
  d12 ;pl9:f1
  d1 ;cw:f1 ph29
  4u do:f1
  20u UNBLKGRAD
  4u pl0:f1
  p11:sp1:f1 ph1:r
  d20
  p16:gp1
  d16
  p11:sp1:f1 ph2:r
  d20
  p16:gp2
  d16
  d12 pl1:f1
```

```
  p1 ph2
  4u BLKGRAD
  go=2 ph31
  30m mc #0 to 2 F0(zd)
exit
ph1=0
ph2=0 2 2 0 1 3 3 1
ph31=0 2 2 0 1 3 3 1
;pl1 : f1 channel - power level for pulse (default)
;pl9 : f1 channel - power level for presaturation
;p1 : f1 channel -  90 degree high power pulse
;d1 : relaxation delay; 1-5 * T1
;d12: delay for power switching                   [20 usec]
;ns: 1 * n, total number of scans: NS * TD0
;$Id: zg,v 1.11 2012/01/31 17:49:32 ber Exp $
```

*2D SMLS-TOCSY with band-selective suppression*

```
; SMLS-TOCSY
;avance-version (19/02/08)
;homonuclear Hartman-Hahn transfer using DIPSI2 sequence
;   for mixing
;phase sensitive
;water suppression using excitation sculpting with gradients
;(use parameterset DIPSI2ESGPPH)
;A.J. Shaka, C.J. Lee & A. Pines, J. Magn. Reson. 77, 274 (1988)
;T.-L. Hwang & A.J. Shaka, J. Magn. Reson.,
;   Series A 112 275-279 (1995)
;$CLASS=HighRes
;$DIM=2D
;$TYPE=
;$SUBTYPE=
```

```
;$COMMENT=
prosol relations=<triple>
#include <Avance.incl>
#include <Delay.incl>
#include <Grad.incl>
"p2=p1*2"
"d11=30m"
"d12=20u"
"d13=4u"

"in0=inf1"
"d0=in0*0.5-p1*4/3.1416"
"TAU=de+p1*2/3.1416+4u"
"FACTOR1=(d9/(p6*115.112))/2"
"l1=FACTOR1*2"
"acqt0=0"
baseopt_echo
1 ze
2 d11
3 d12 ;pl32:f1
  d1 ;cw:f1 ph29
  d13 do:f1
  20u UNBLKGRAD
  4u pl0:f1
  p13:sp3:f1 ph1:r
  d20
  p16:gp1
```

d16

p13:sp3:f1 ph1:r

d20

p16:gp2

d16

d12 pl1:f1

p1 ph2

d0

p1 ph3

;50u UNBLKGRAD

p16:gp3

d16 pl10:f1

;begin DIPSI2

4 p6*3.556 ph23

p6*4.556 ph25

p6*3.222 ph23

p6*3.167 ph25

p6*0.333 ph23

p6*2.722 ph25

p6*4.167 ph23

p6*2.944 ph25

p6*4.111 ph23

p6*3.556 ph25

p6*4.556 ph23

p6*3.222 ph25

p6*3.167 ph23

```
p6*0.333 ph25
p6*2.722 ph23
p6*4.167 ph25
p6*2.944 ph23
p6*4.111 ph25
p6*3.556 ph25
p6*4.556 ph23
p6*3.222 ph25
p6*3.167 ph23
p6*0.333 ph25
p6*2.722 ph23
p6*4.167 ph25
p6*2.944 ph23
p6*4.111 ph25
p6*3.556 ph23
p6*4.556 ph25
p6*3.222 ph23
p6*3.167 ph25
p6*0.333 ph23
p6*2.722 ph25
p6*4.167 ph23
p6*2.944 ph25
p6*4.111 ph23
lo to 4 times l1                                    ;end DIPSI
4u
p16:gp4
d16 pl1:f1
```

```
  p1 ph4
  p16:gp5
  d16 pl0:f1
  (p12:sp1 ph5:r):f1
  4u
  d12 pl1:f1
  p2 ph6
  4u
  p16:gp5
  d16
  TAU
  p16:gp6
  d16 pl0:f1
  (p12:sp1 ph7:r):f1
  4u
  d12 pl1:f1
  p2 ph8
  4u
  p16:gp6
  d16
  4u BLKGRAD
  go=2 ph31
  d11 mc #0 to 2 F1PH(calph(ph1, +90) & calph(ph29, +90), caldel(d0, +in0))
exit
ph1=0
ph2=0 2
ph3=0 0 0 0 2 2 2 2
```

ph4=0 0 0 0 0 0 0 0 2 2 2 2 2 2 2 2

ph5=0 0 1 1

ph6=2 2 3 3

ph7=0 0 0 0 1 1 1 1

ph8=2 2 2 2 3 3 3 3

ph23=3

ph25=1

;ph29=0

ph31=0 2 2 0 0 2 2 0 2 0 0 2 2 0 0 2

;pl0 : 0W

;pl1 : f1 channel - power level for pulse (default)

;pl10: f1 channel - power level for TOCSY-spinlock

;pl32: f1 channel - power level for low power presaturation

;sp1 : f1 channel - shaped pulse 180 degree

;p1 : f1 channel -  90 degree high power pulse

;p2 : f1 channel - 180 degree high power pulse

;p6 : f1 channel -  90 degree low power pulse

;p12: f1 channel - 180 degree shaped pulse (Squa100.1000)   [2 msec]

;p16: homospoil/gradient pulse

;d0 : incremented delay (2D)

;d1 : relaxation delay; 1-5 * T1

;d9 : TOCSY mixing time

;d11: delay for disk I/O                  [30 msec]

;d12: delay for power switching                  [20 usec]

;d13: short delay                  [4 usec]

;d16: delay for homospoil/gradient recovery

;l1: loop for DIPSI cycle: ((p6*115.112) * l1) = mixing time

;inf1: 1/SW = 2 * DW

;in0: 1/(1 * SW) = 2 * DW

;nd0: 1

;ns: 8 * n

;ds: 16

;td1: number of experiments

;FnMODE: States-TPPI, TPPI, States or QSEQ

;use gradient ratio:  gp 1 : gp 2 : gp 3 : gp 4

;  1 :  3 :  31 :  11

;for z-only gradients:

;gpz1: 1%

;gpz2: 3%

;gpz3: 31%

;gpz4: 11%

;use gradient files:

;gpnam1: SMSQ10.100

;gpnam2: SMSQ10.100

;gpnam3: SMSQ10.100

;gpnam4: SMSQ10.100

;set pl32 to 0W when presaturation is not required

;  use pl1 + 75 to 80dB to reduce radiation damping

;Processing

;PHC0(F1): 90

;PHC1(F1): -180

;FCOR(F1): 1

;$Id: dipsi2esgpph,v 1.15 2019/02/11 14:04:14 ber Exp $