

# Quantitative NMR using cryo probe

Spectral Service



Laboratorium  
für Auftragsanalytik GmbH



## 100<sup>th</sup> AOCS Annual Meeting

Quantitative  $^{31}\text{P}$ -NMR mit cQNP at 600 MHz

Orlando, FL, May 2009

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# Lipids

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## Sources

Animal

Milk

Egg

Fish

Krill

.....

Vegetable

Soy

Rape

Olive

Sunflower

.....

## Classes

Triglycerides

Phospholipids

Glycolipides

(Sterols)

# Alternative: qNMR of Lipids using cryo probe

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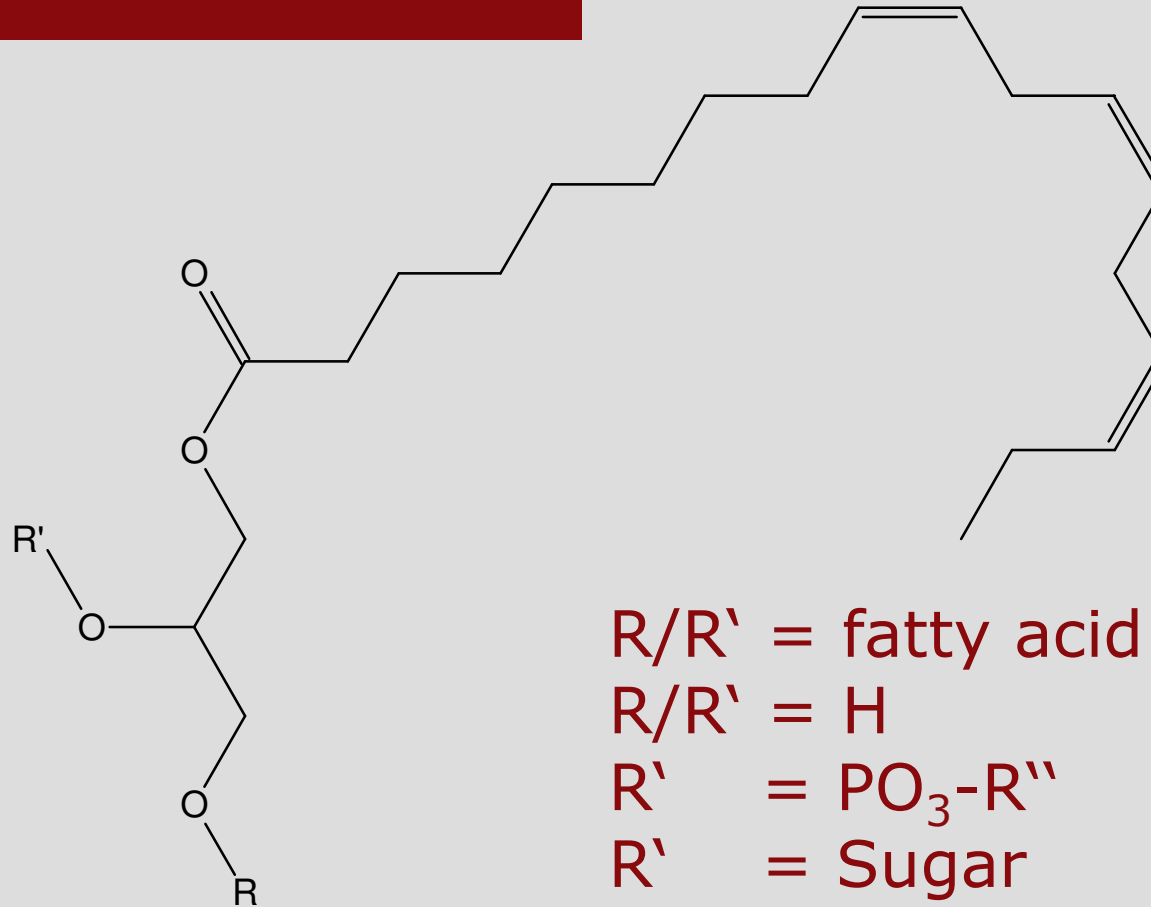
Parameter	Analytical Methode	Alternative!
Iodine Value	Titration, unspecific	1H NMR
Peroxide Value Oil	Titration, unspecific	1H NMR
Peroxide Value Phospholipid's	???	1H NMR
Acid value	Titration, unspecific	1H NMR
Amount of unsaturated FA	GC specific with standards	1H NMR
Amount of 18:1, 18:2, 18:3	GC specific with standards	1H NMR
Amount of PUFA (w-3)	GC specific with standards	1H NMR
Amount of Steroles	GC specific with standards	1H NMR
Amount of Phospholipids	HPLC-LSD specific with standards	31P NMR
Phospholipid Distribution	HPLC-LSD specific with standards	31P NMR
Fatty acid distribution in SN1/SN2	Enzymatic/GC	13C NMR
Fatty acid distribution in w-n	GC specific with standards	13C NMR
Mono-Di Triglyceride	????	13C NMR
Animal/Vegetable origin	Combination of several	NMR
Synthetic/semisynthetic e.g. PS	Mostly no standard	NMR

# qNMR of Lipids using cryo probe

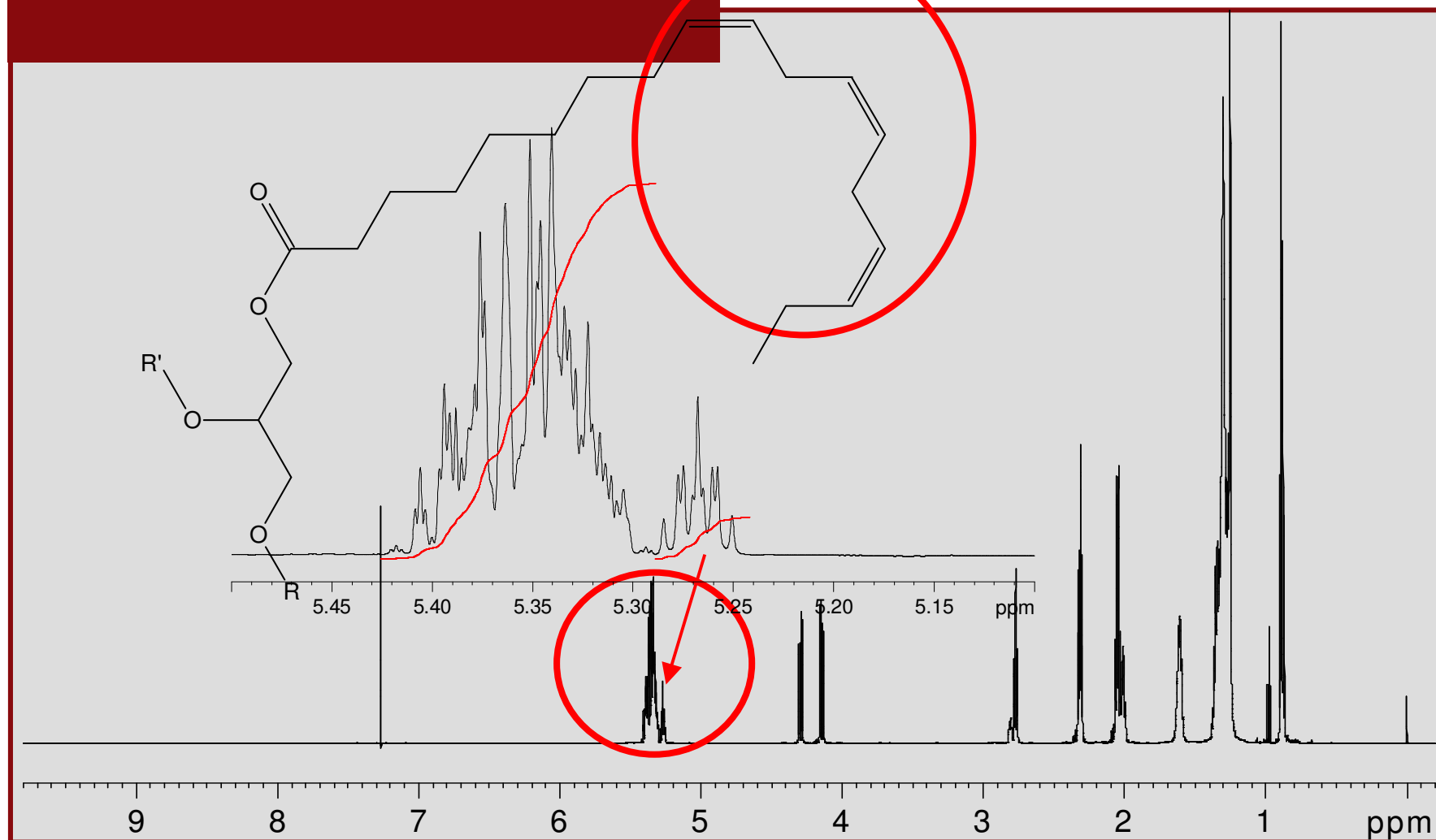
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# $^1\text{H}$ NMR of Soybean Oil

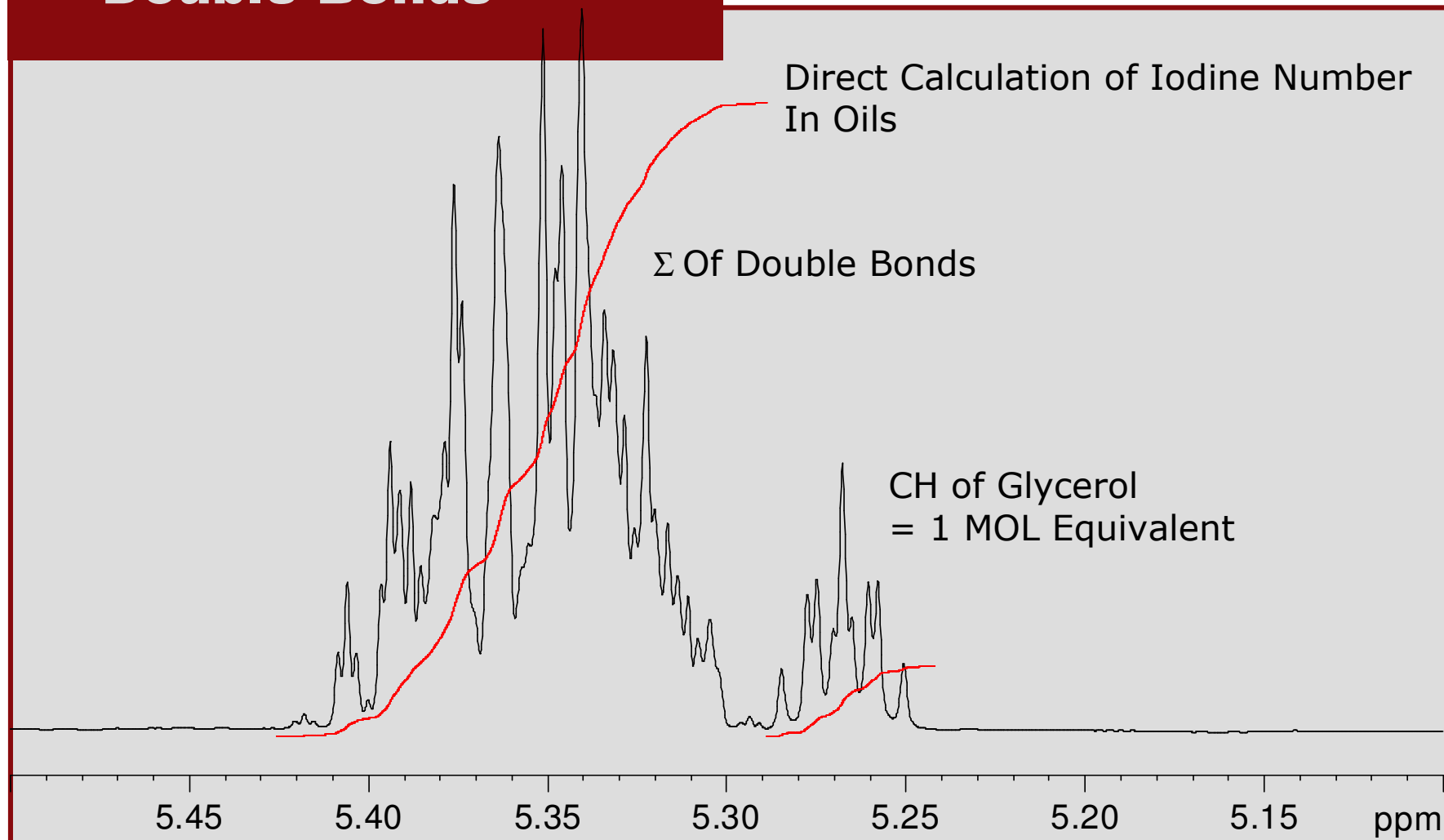


# <sup>1</sup>H NMR of Soybean Oil Double Bonds

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# <sup>1</sup>H NMR of Soybean PC Double Bonds

Spectral Service

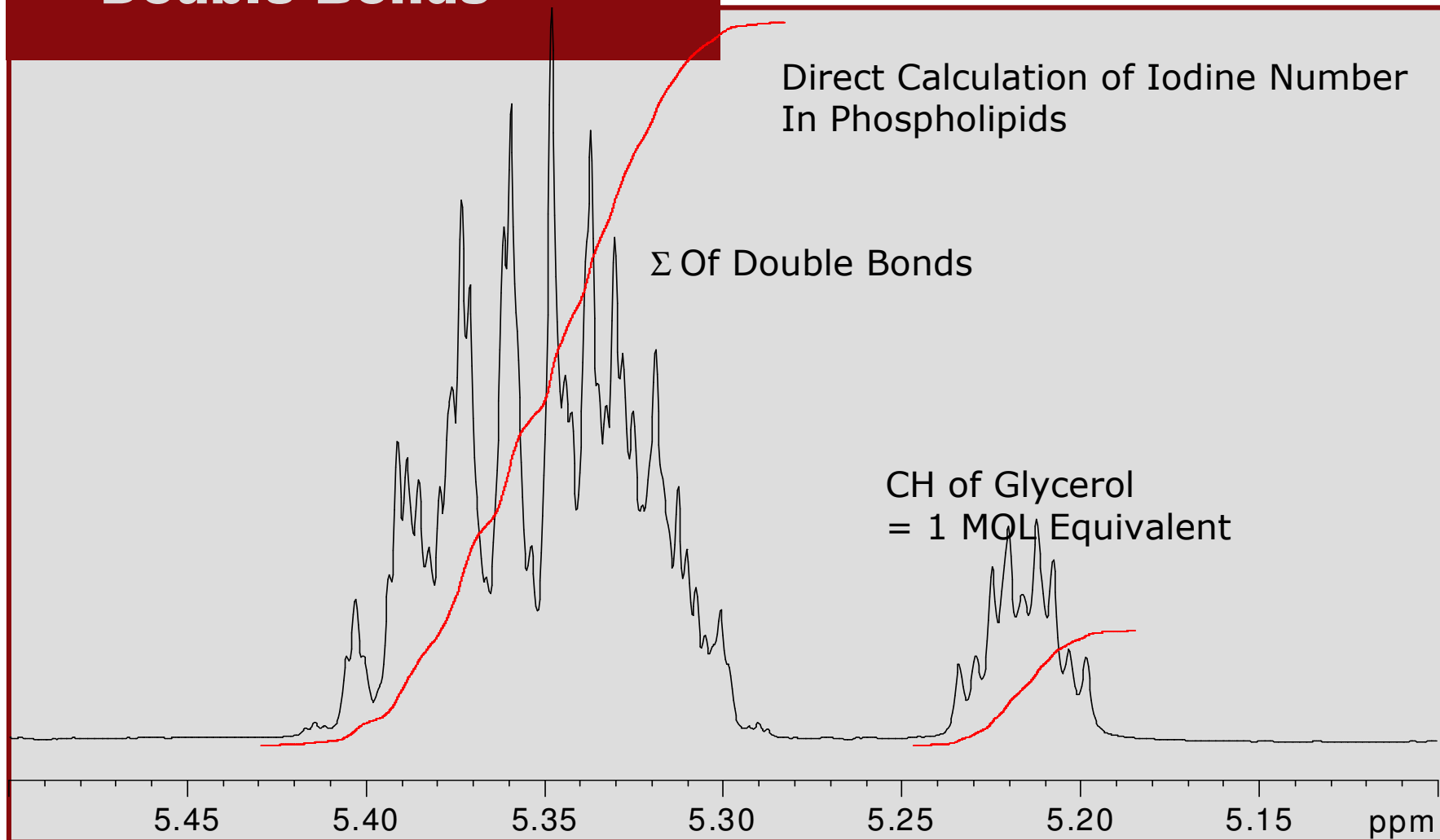


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Direct Calculation of Iodine Number In Phospholipids

$\Sigma$  Of Double Bonds

CH of Glycerol = 1 MOL Equivalent



# <sup>1</sup>H NMR of Soybean Oil + PC Double Bonds

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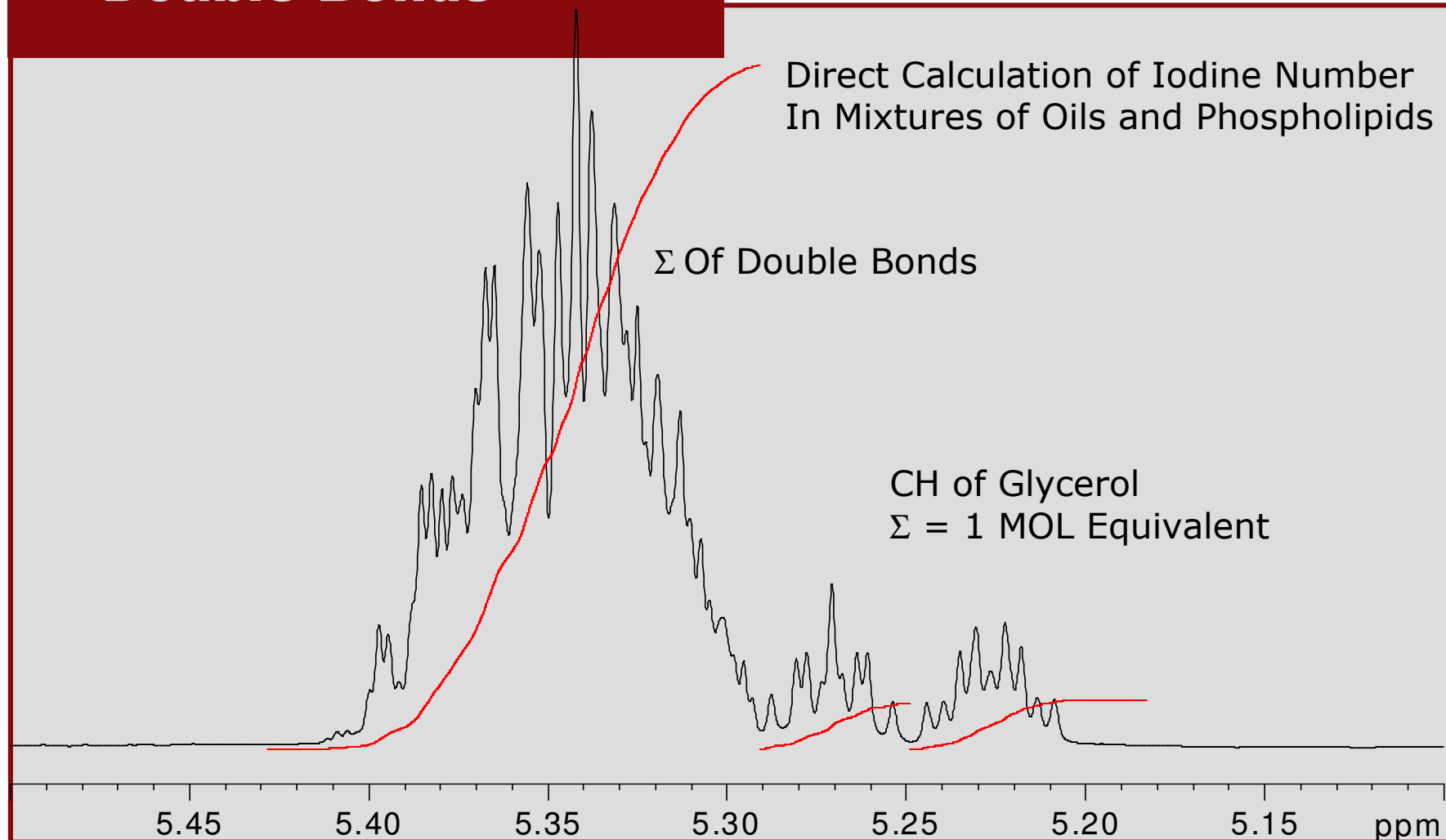


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Direct Calculation of Iodine Number In Mixtures of Oils and Phospholipids

$\Sigma$  Of Double Bonds

CH of Glycerol  
 $\Sigma = 1$  MOL Equivalent



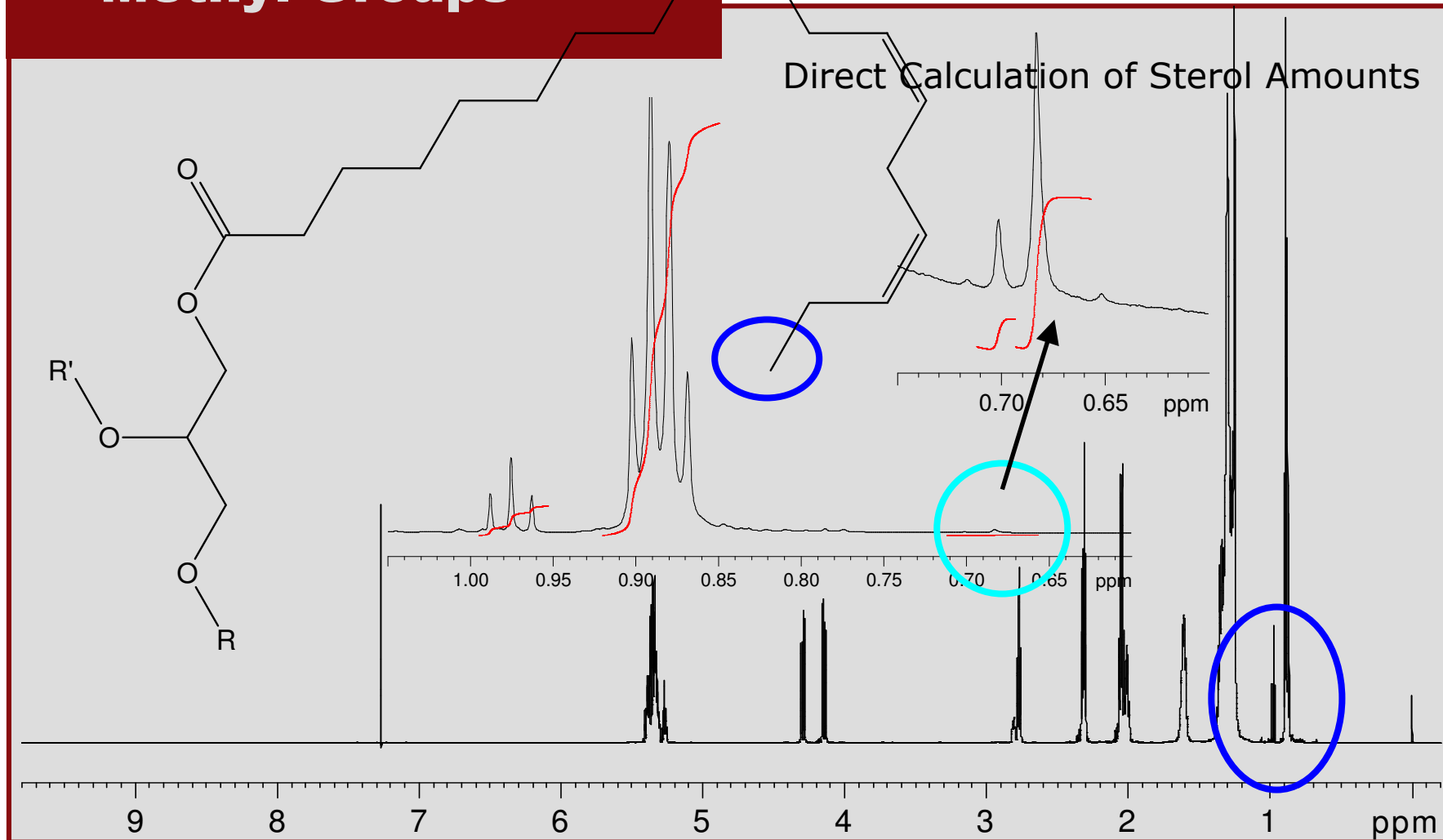


# <sup>1</sup>H NMR of Soybean Oil Methyl Groups

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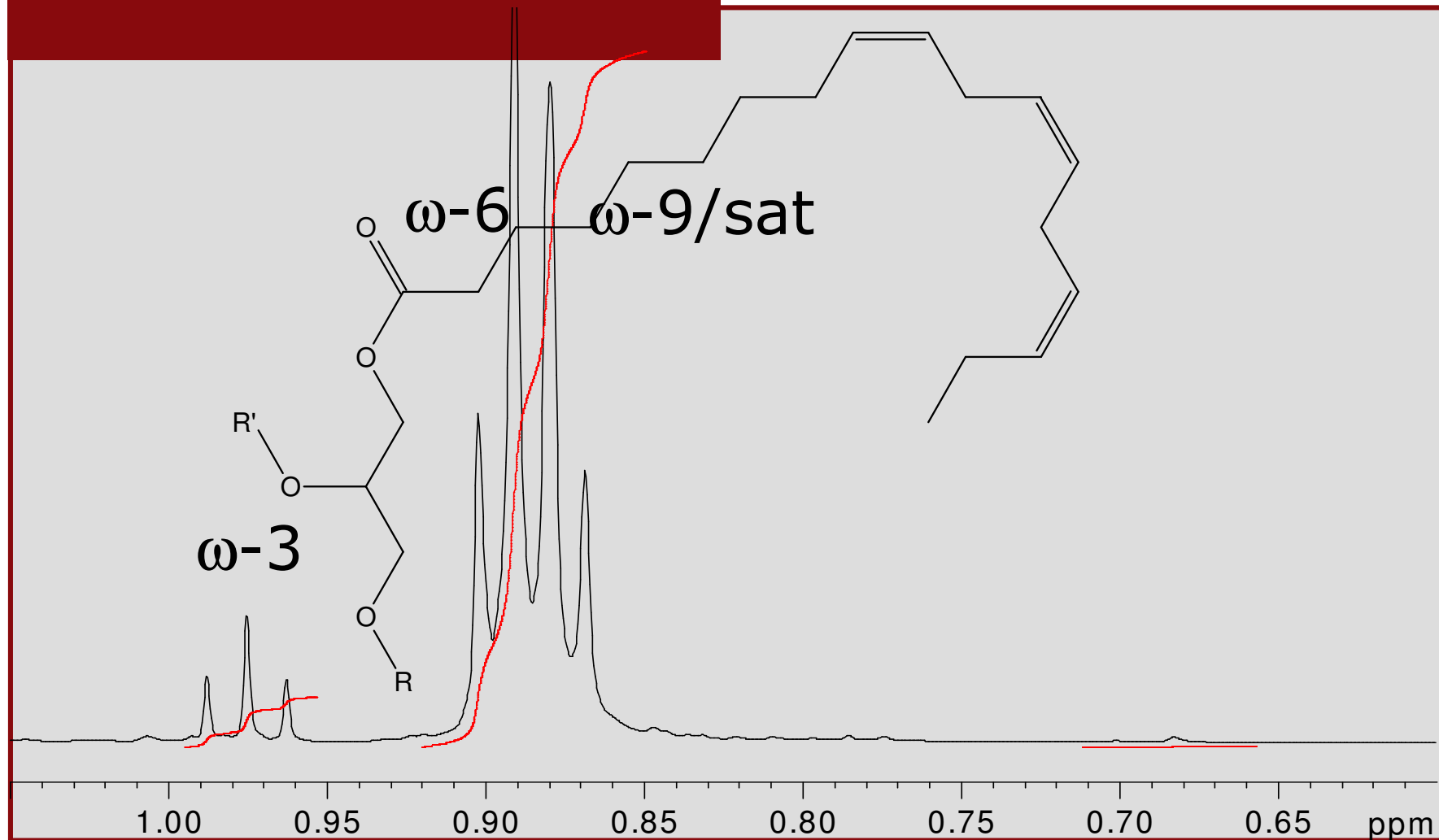


# <sup>1</sup>H NMR of Soybean Oil

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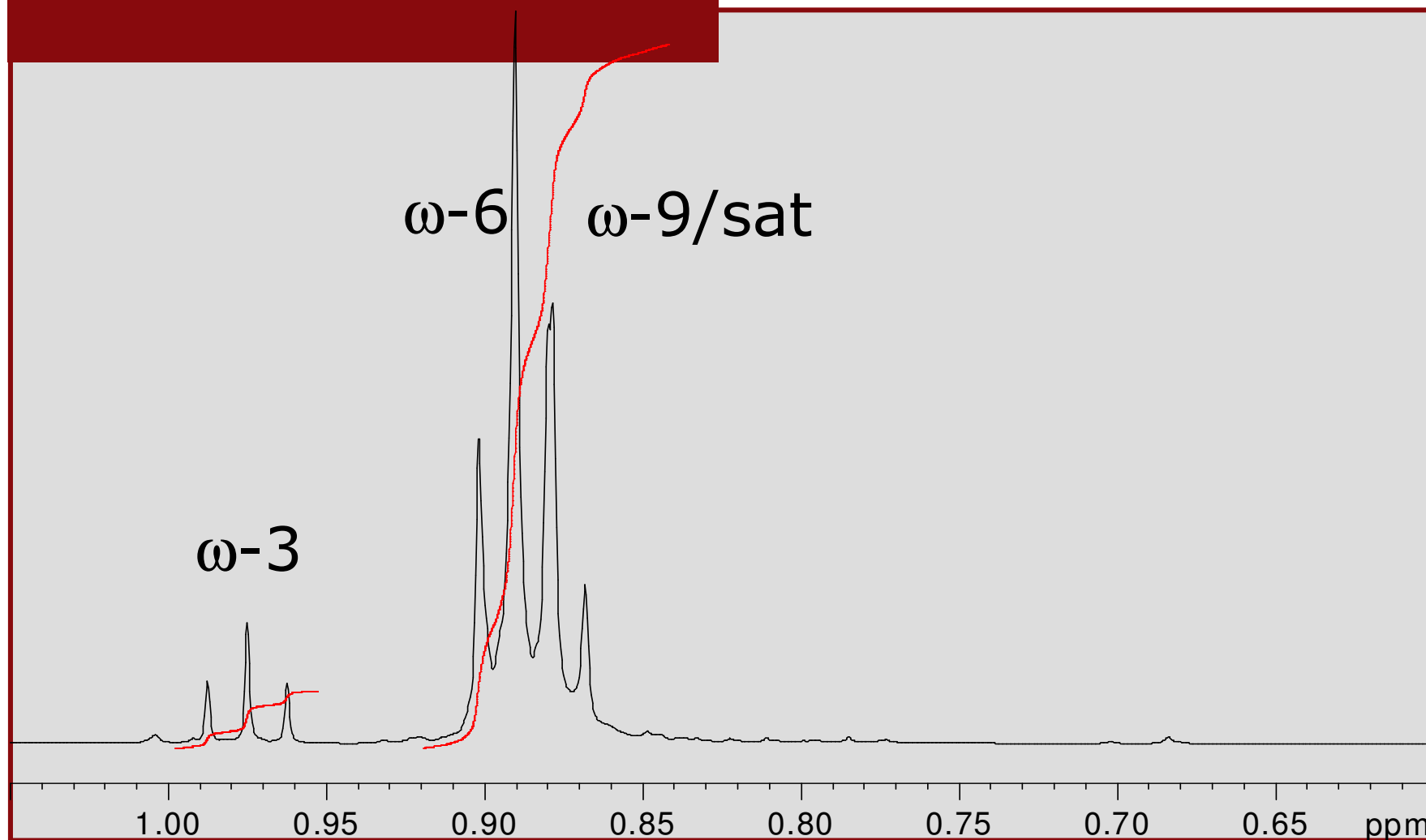


# $^1\text{H}$ NMR of Soybean PC

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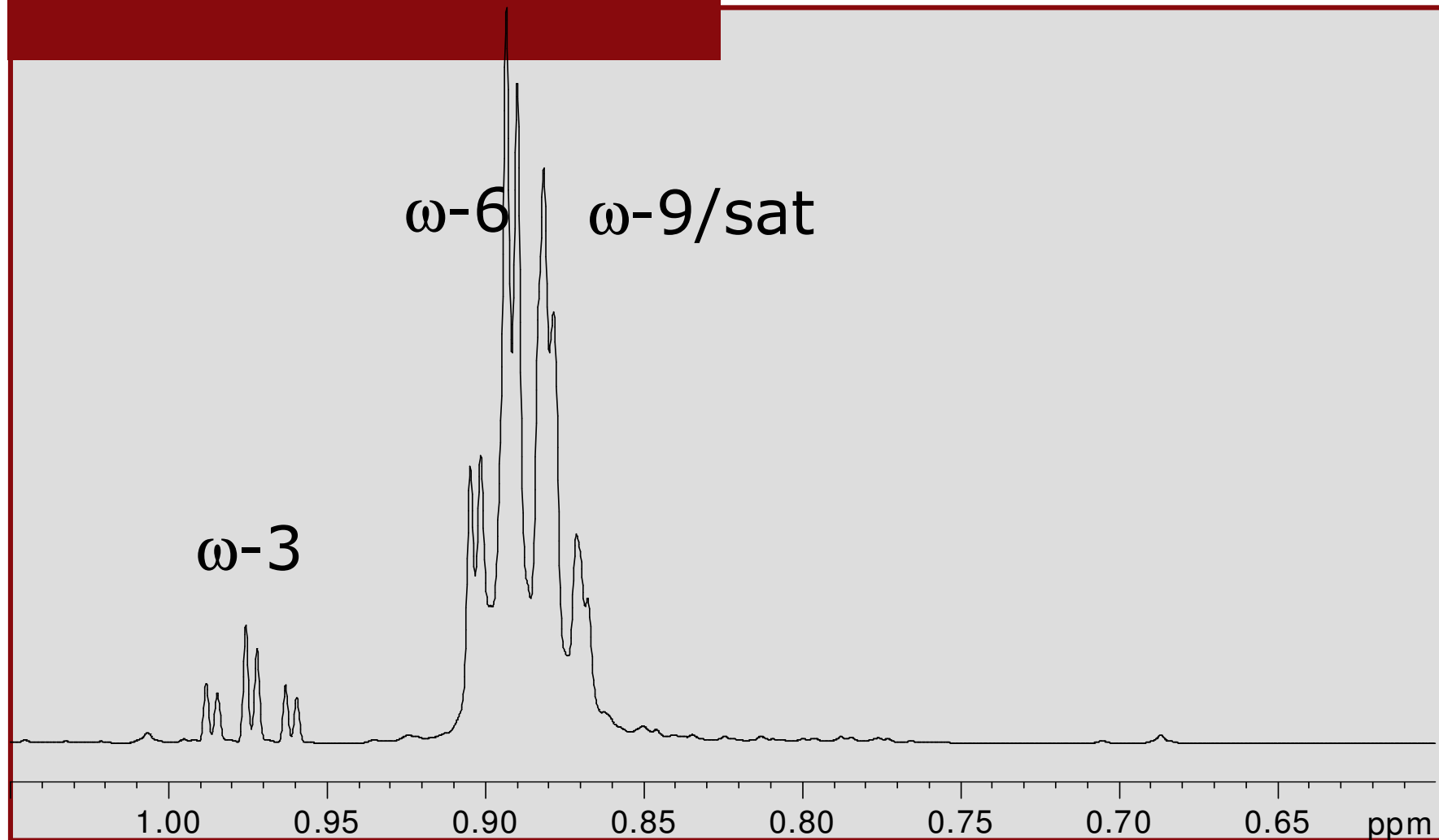


# $^1\text{H}$ NMR of Soybean Oil + PC

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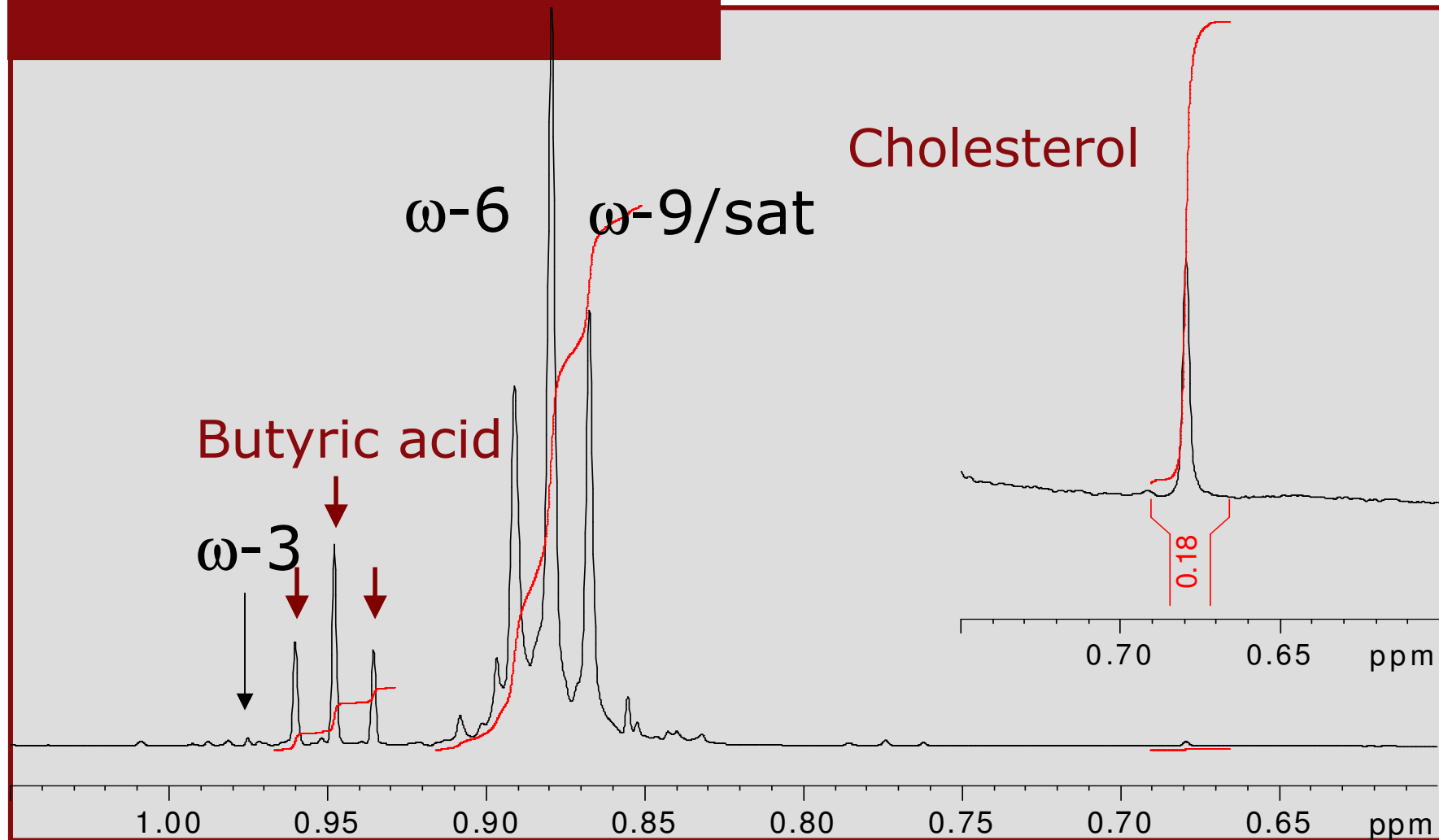


# $^1\text{H}$ NMR of Milk

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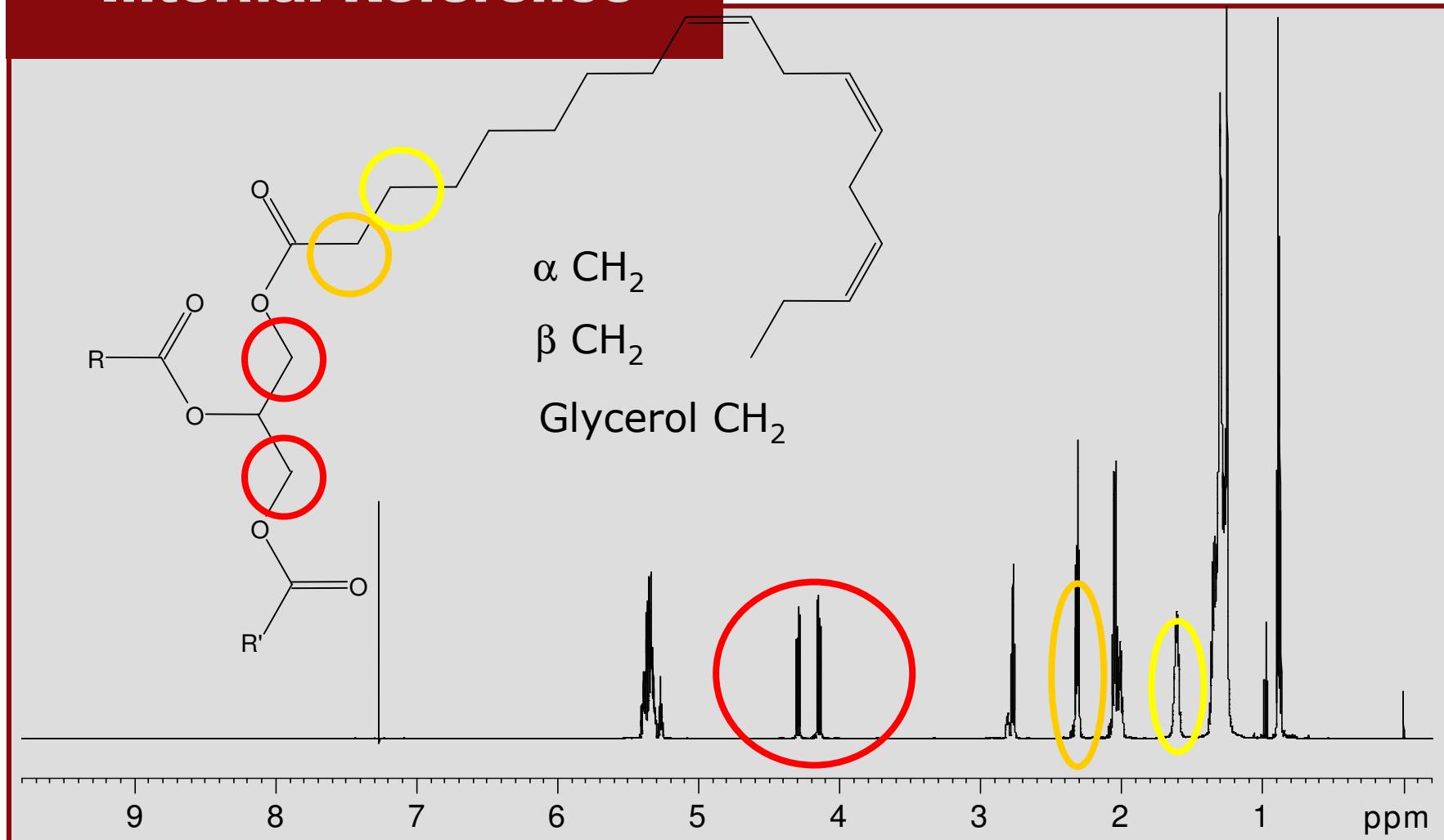


# <sup>1</sup>H NMR of Soybean Oil Internal Reference

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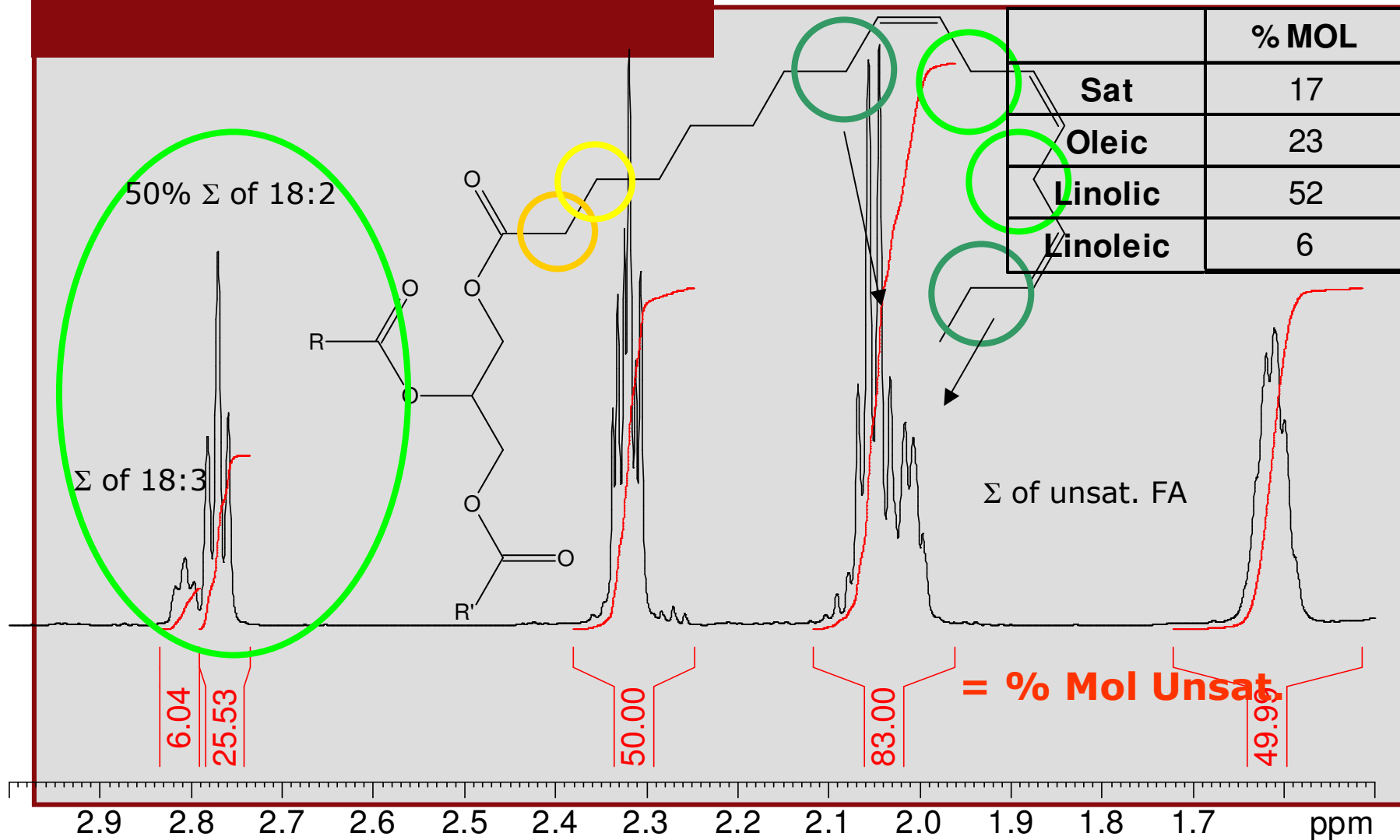


# <sup>1</sup>H NMR of Soybean Oil

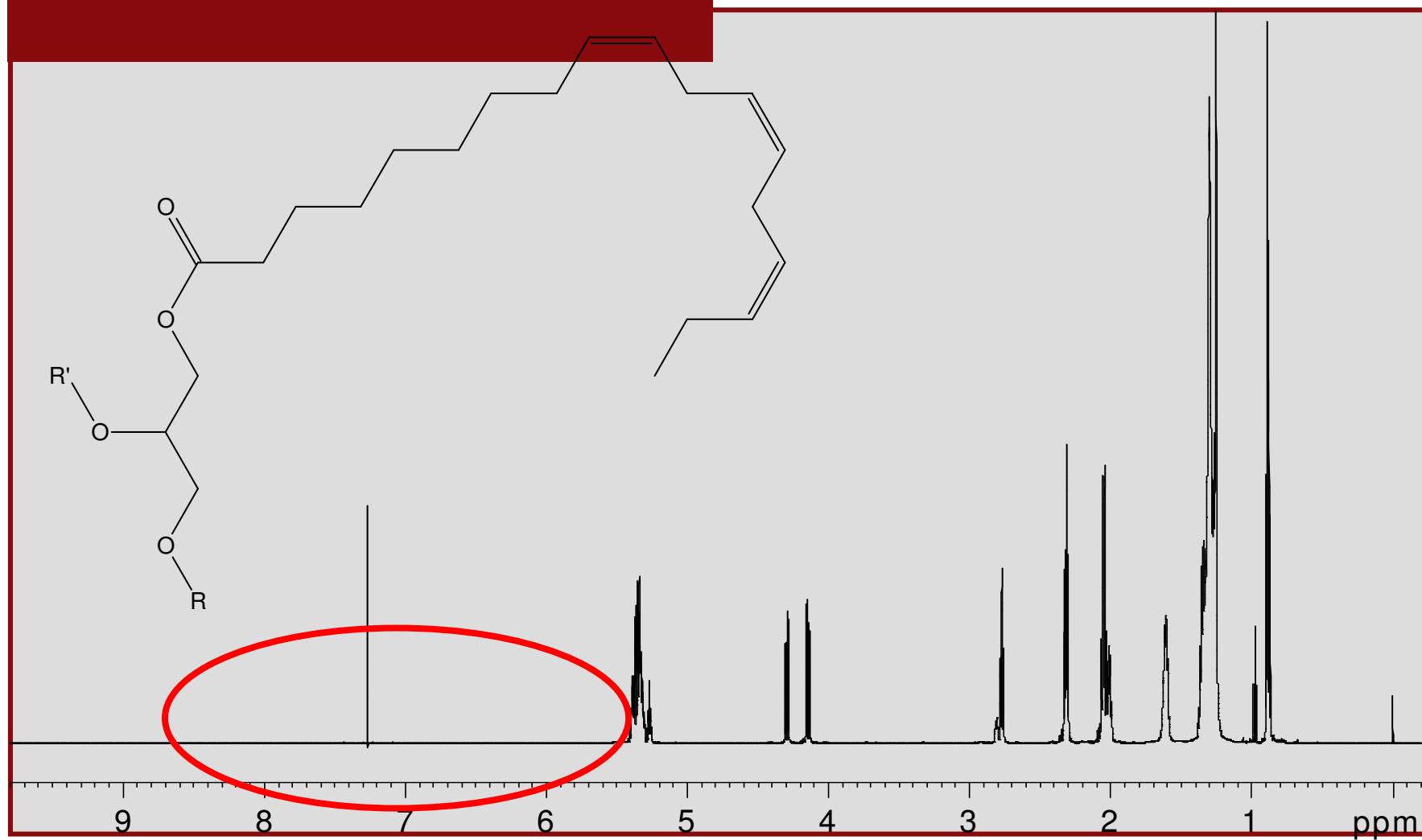
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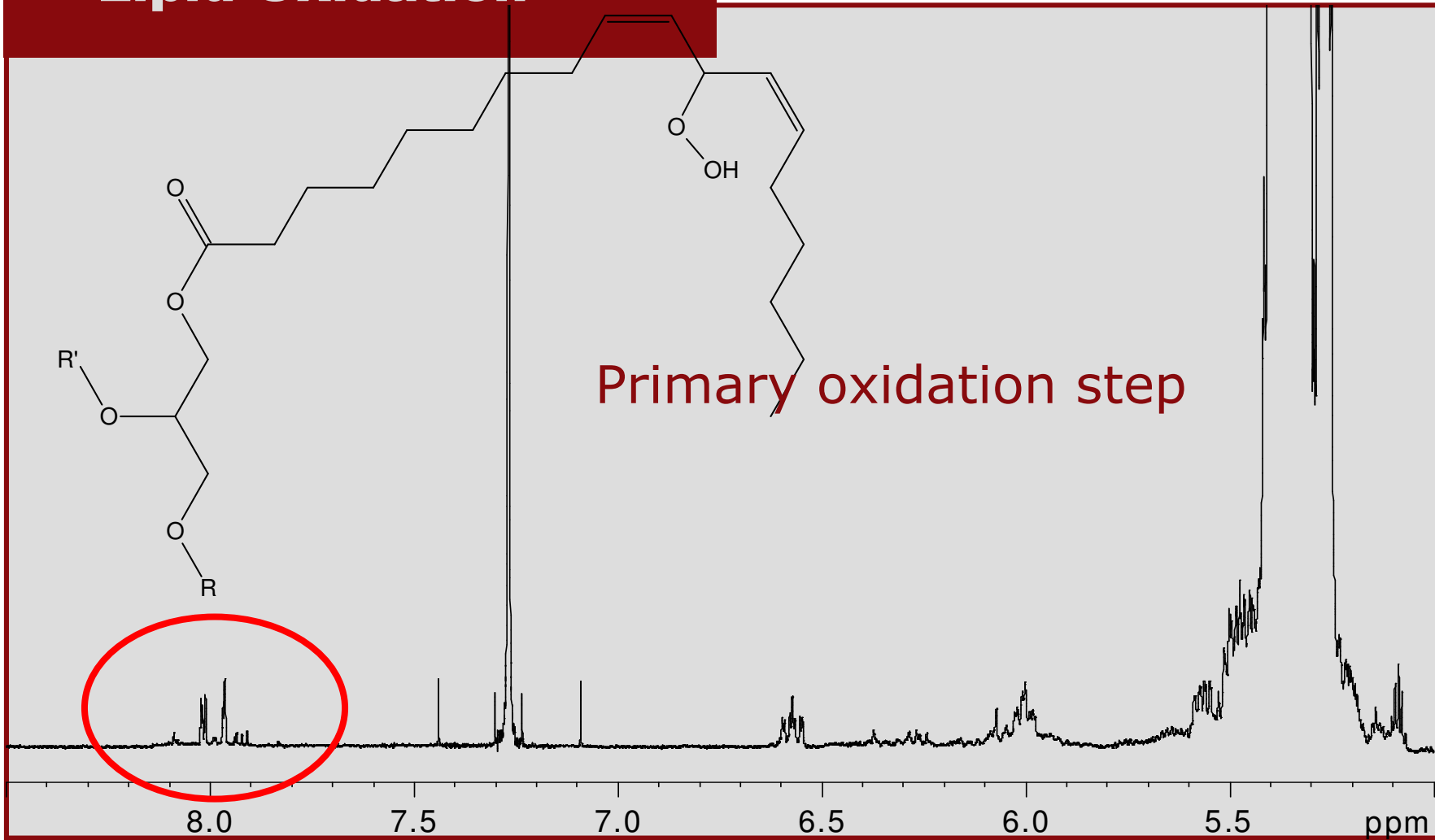


# $^1\text{H}$ NMR of Soybean Oil

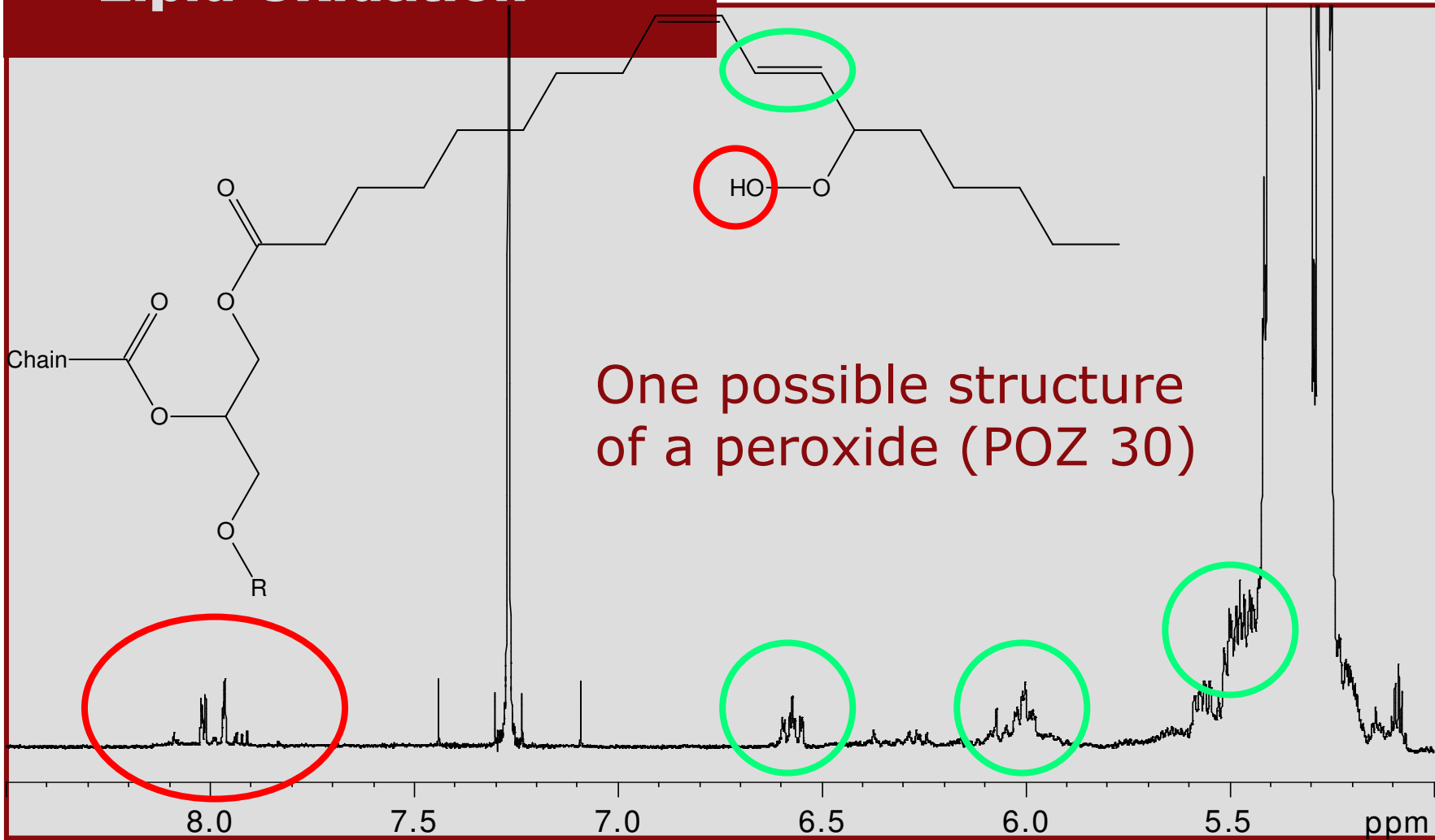




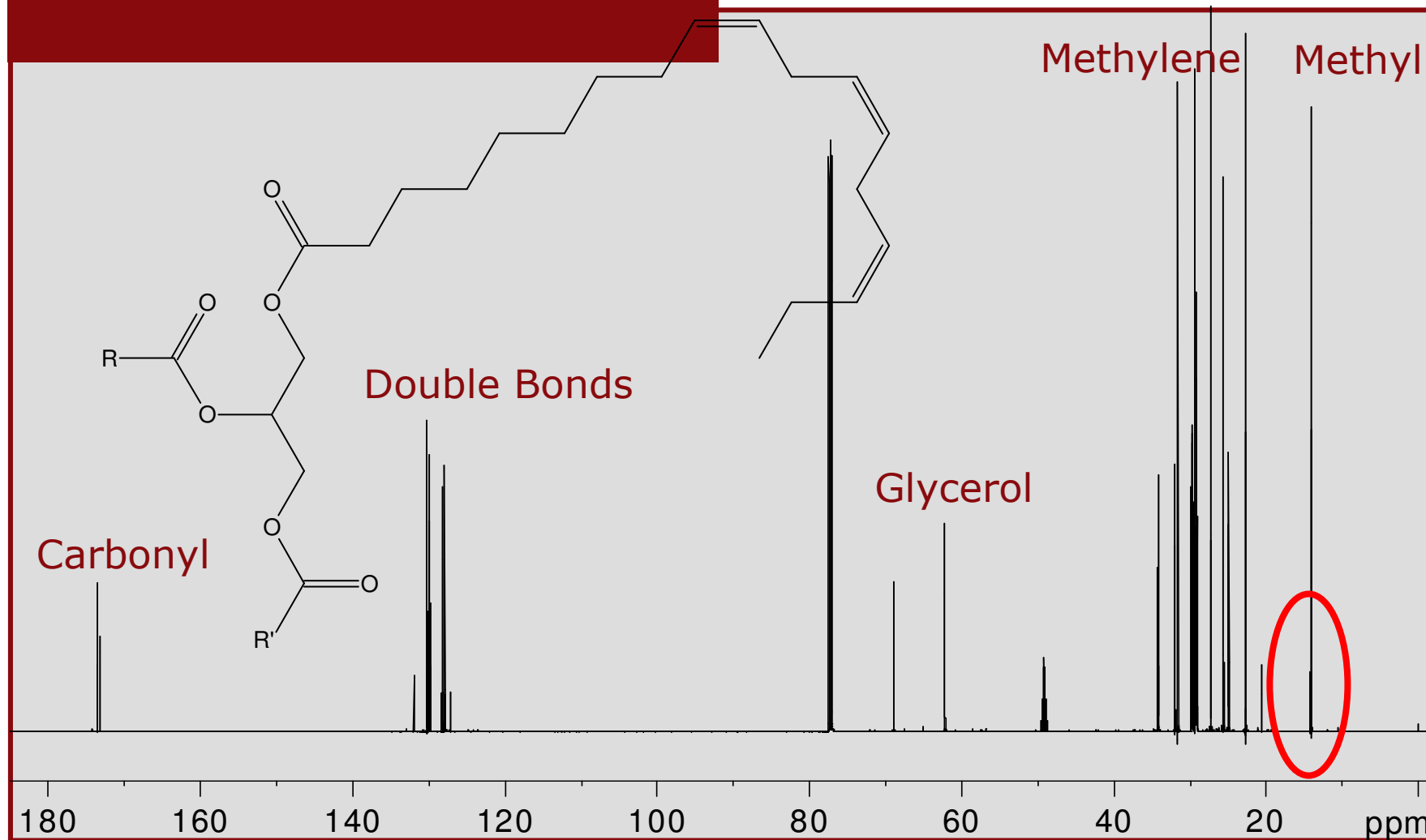
# **<sup>1</sup>H NMR of Soybean Oil Lipid Oxidation**



# <sup>1</sup>H NMR of Soybean Oil Lipid Oxidation



# <sup>13</sup>C NMR of Soybean Oil

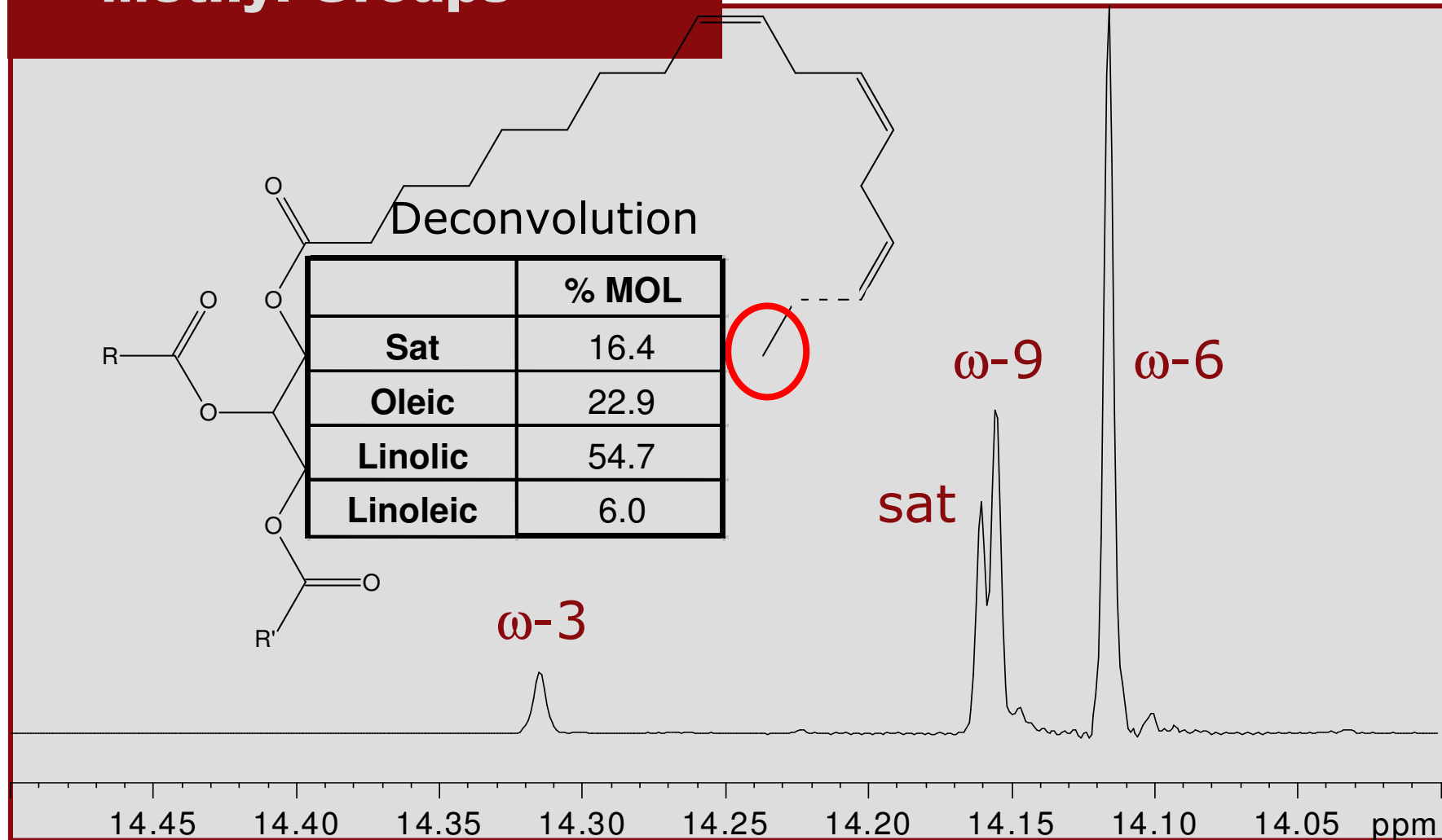


# <sup>13</sup>C NMR of Soybean Oil Methyl Groups

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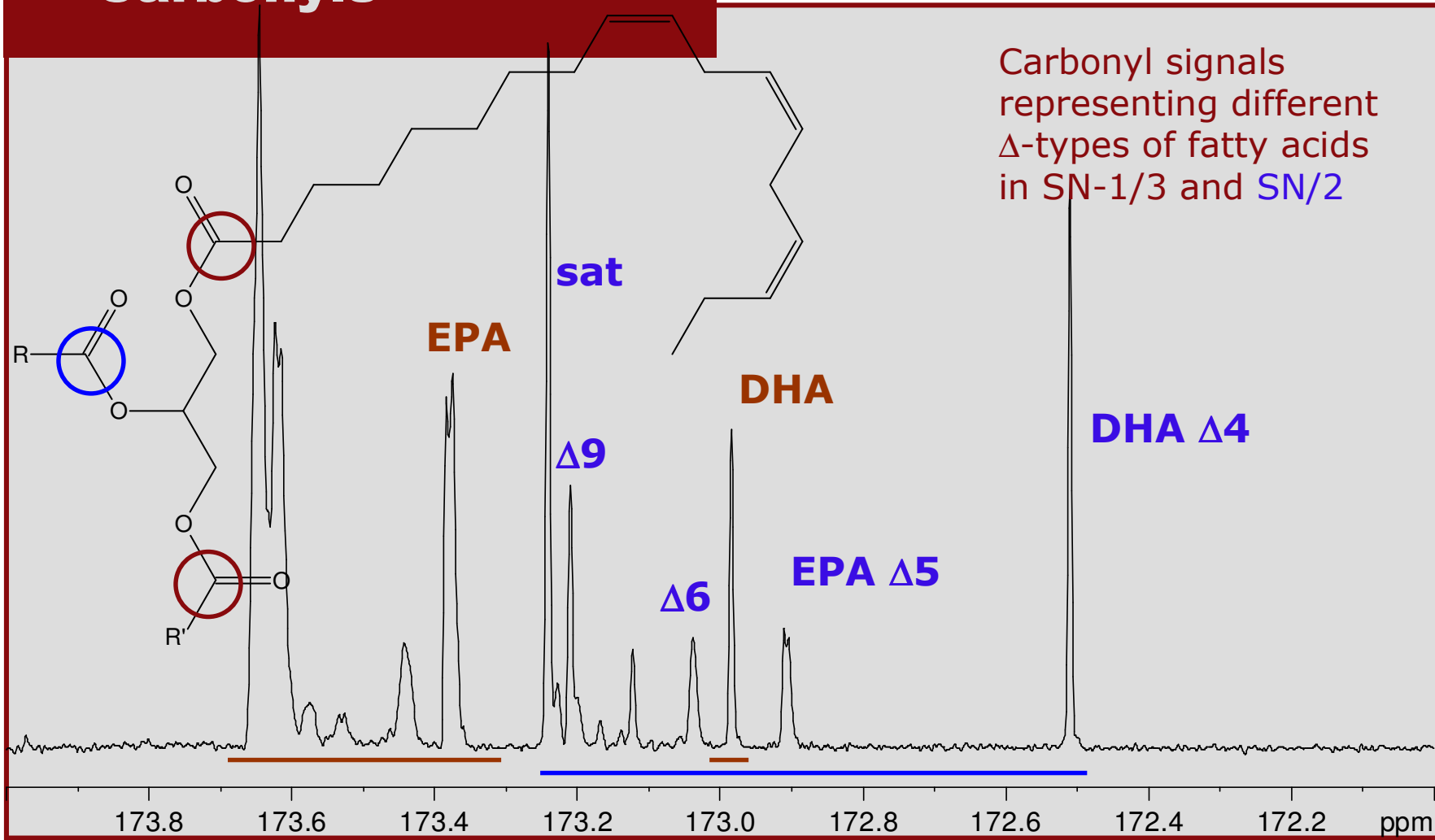
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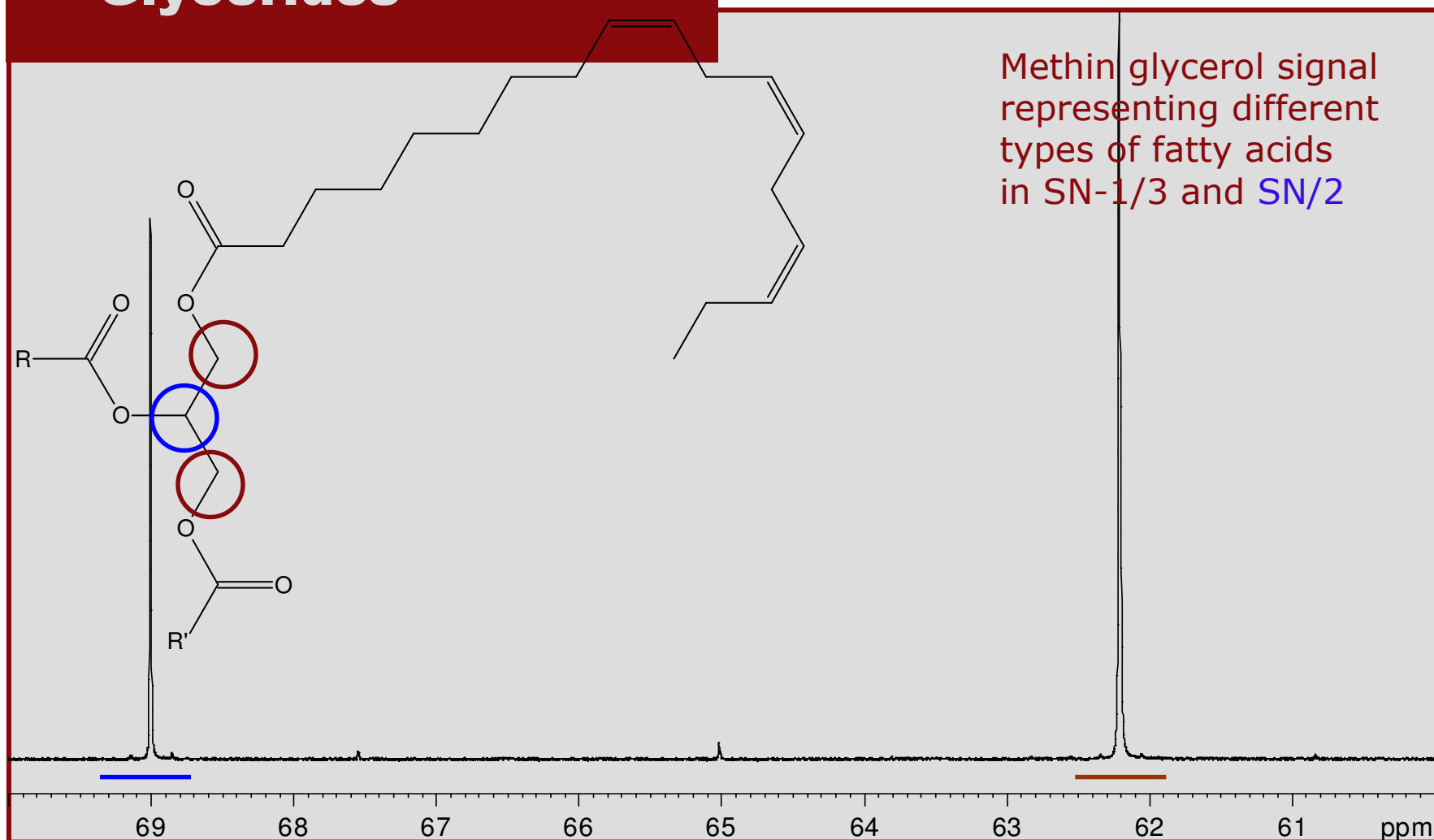




# <sup>13</sup>C NMR of Salmon Oil Carbonyls



# <sup>13</sup>C NMR of Soybean Oil Glycerides

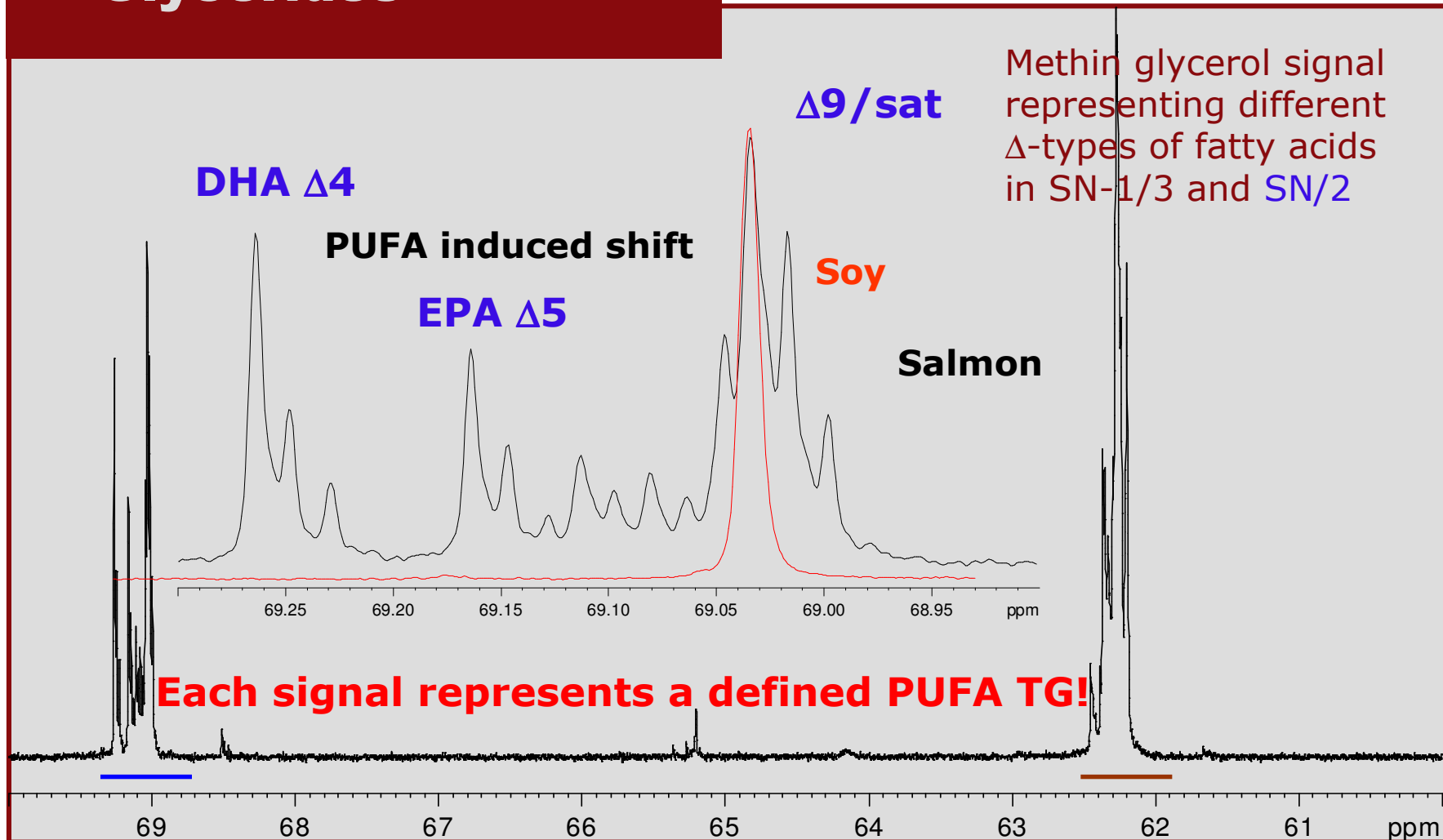


# <sup>13</sup>C NMR of Salmon Oil Glycerides

Spectral Service



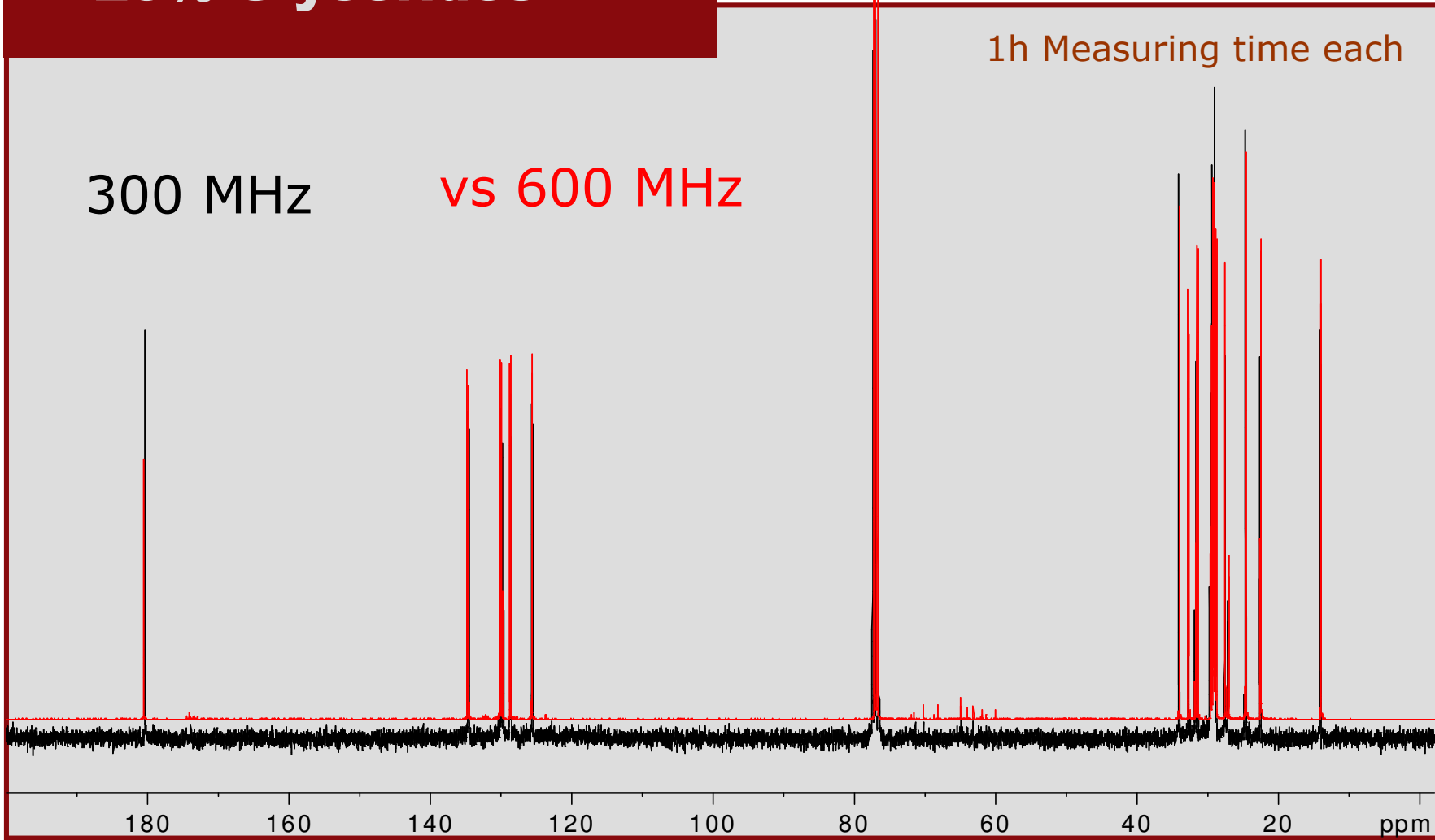
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**<sup>13</sup>C NMR of  
80% CLA Acid  
20% Glycerides**





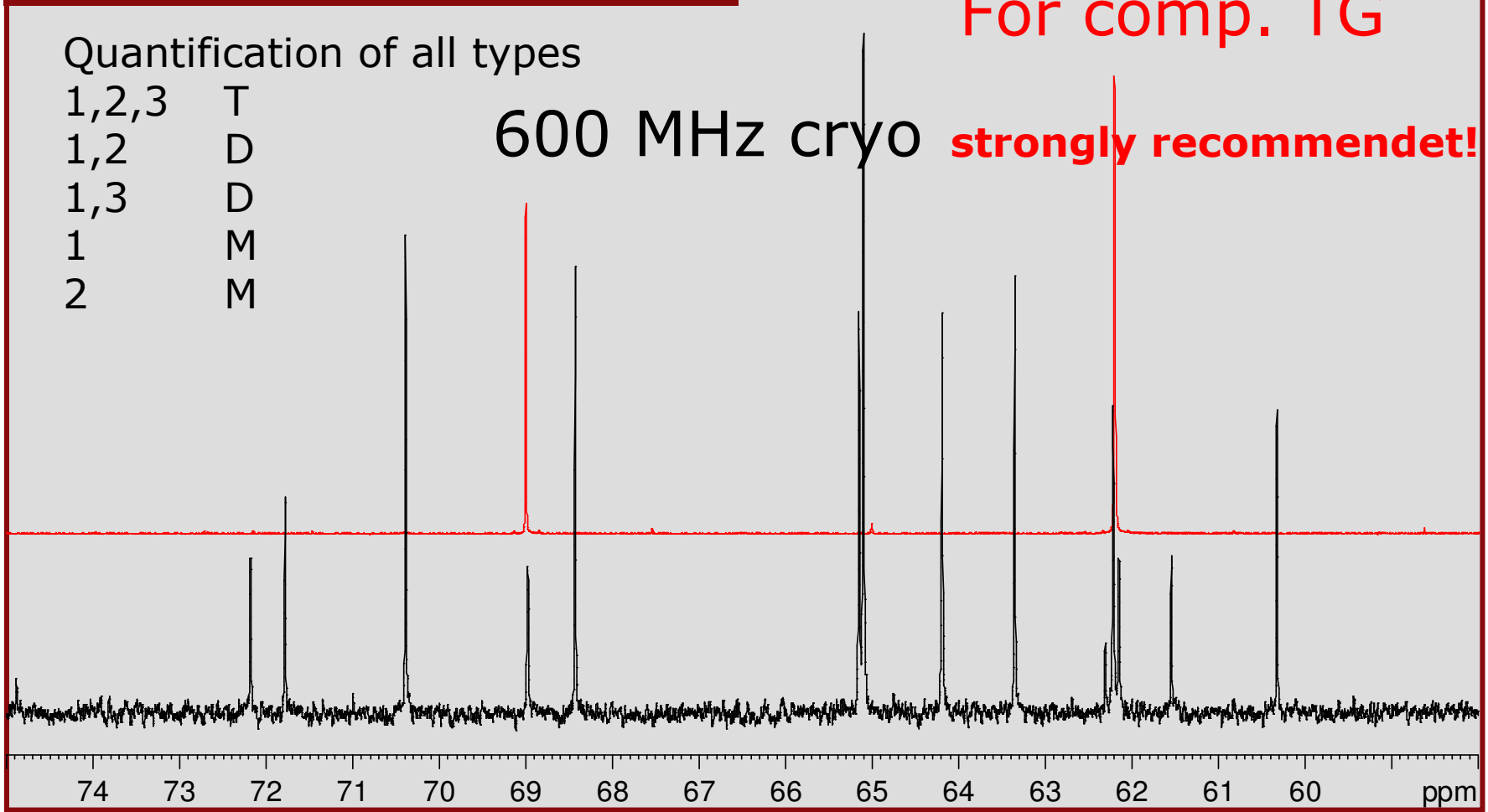
# <sup>13</sup>C NMR of 80% CLA Acid 20% Glycerides

Quantification of all types

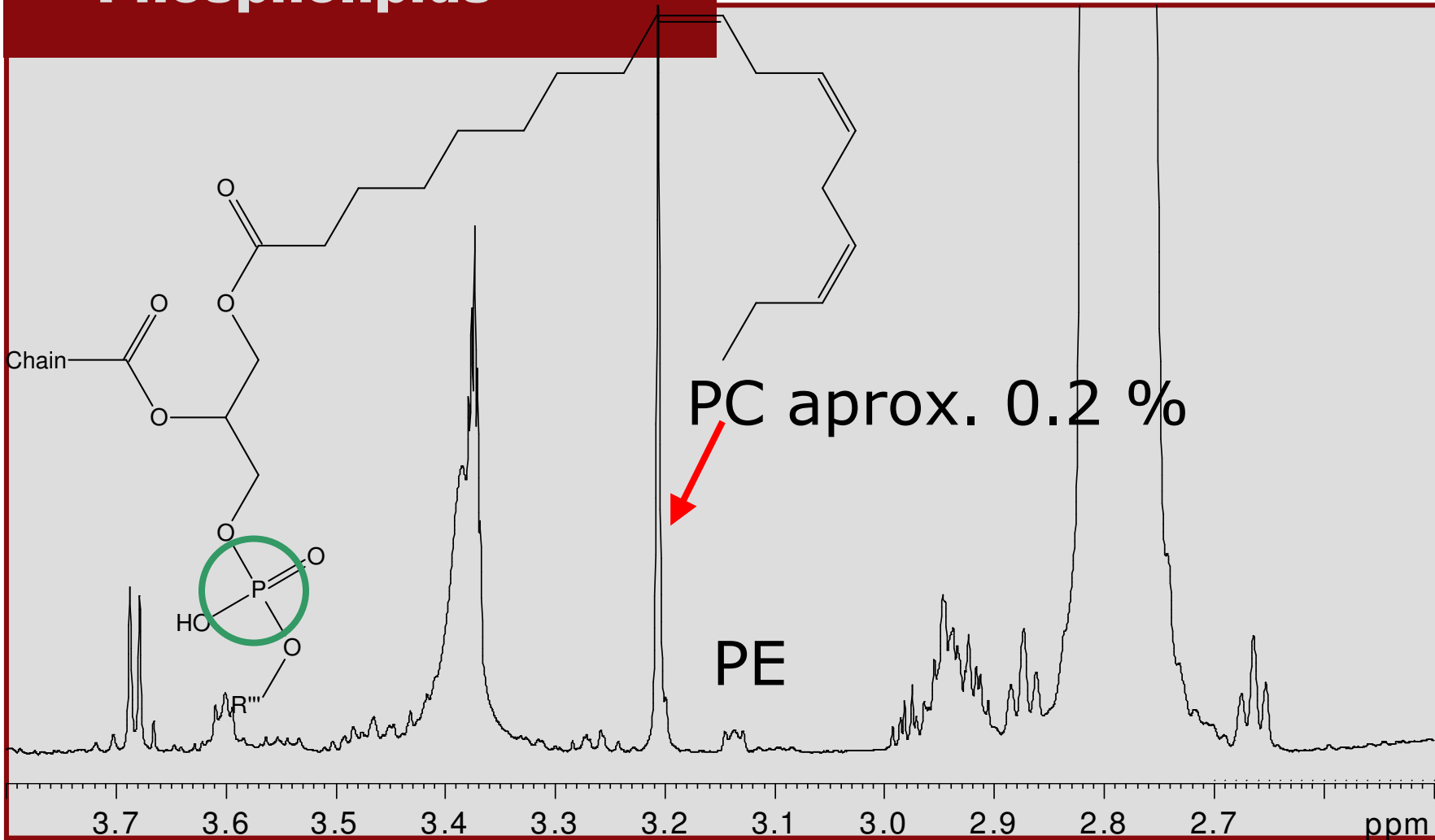
- 1,2,3 T
- 1,2 D
- 1,3 D
- 1 M
- 2 M

600 MHz cryo **strongly recommendet!**

For comp. TG



# <sup>1</sup>H NMR of Soybean Oil Phospholipids

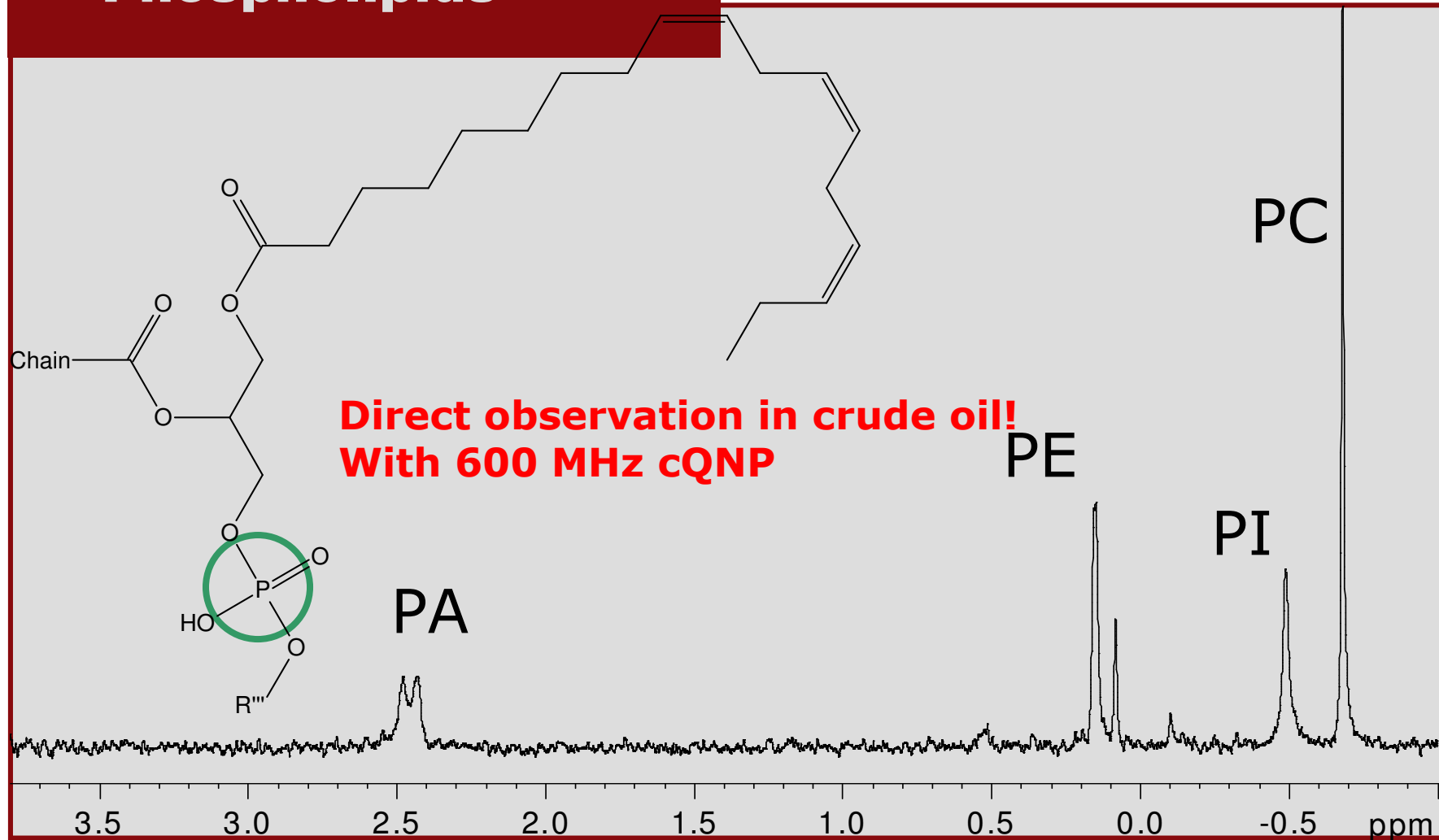


# **$^{31}\text{P}$ NMR of Soybean Oil Phospholipids**

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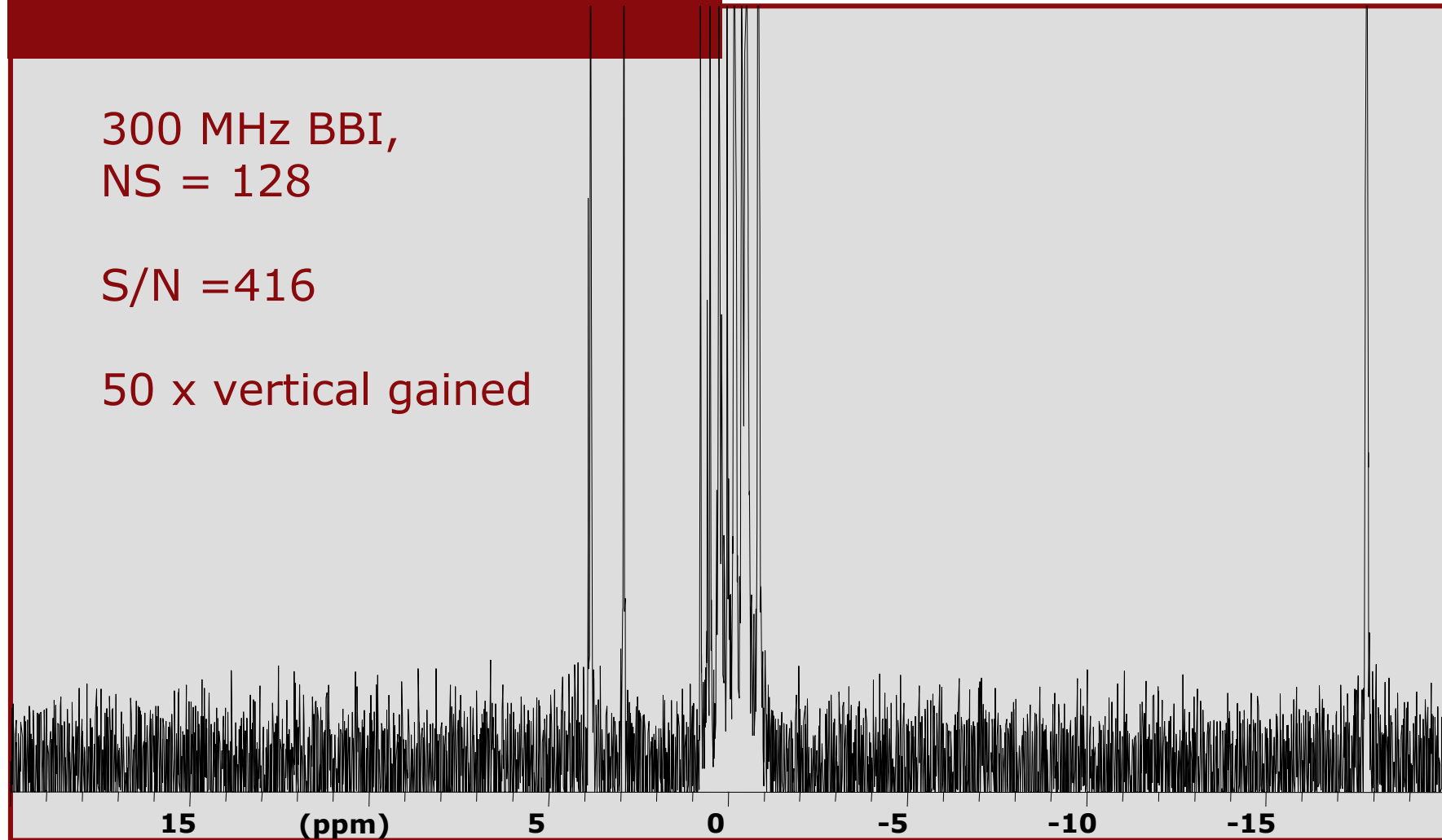


# **$^{31}\text{P}$ NMR of Phospholipids**

300 MHz BBI,  
NS = 128

S/N = 416

50 x vertical gained



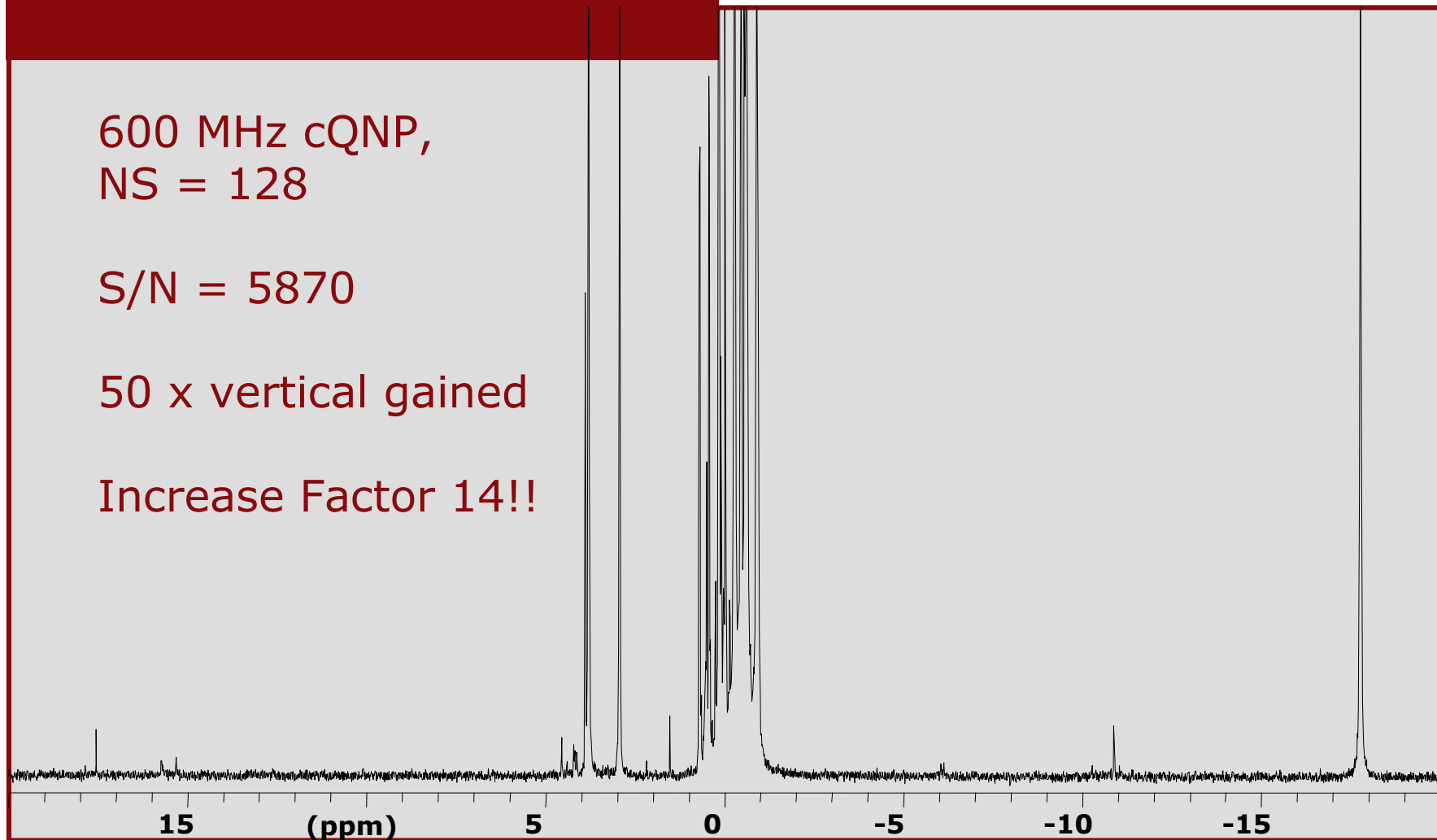
# **$^{31}\text{P}$ NMR of Phospholipids**

600 MHz cQNP,  
NS = 128

S/N = 5870

50 x vertical gained

Increase Factor 14!!

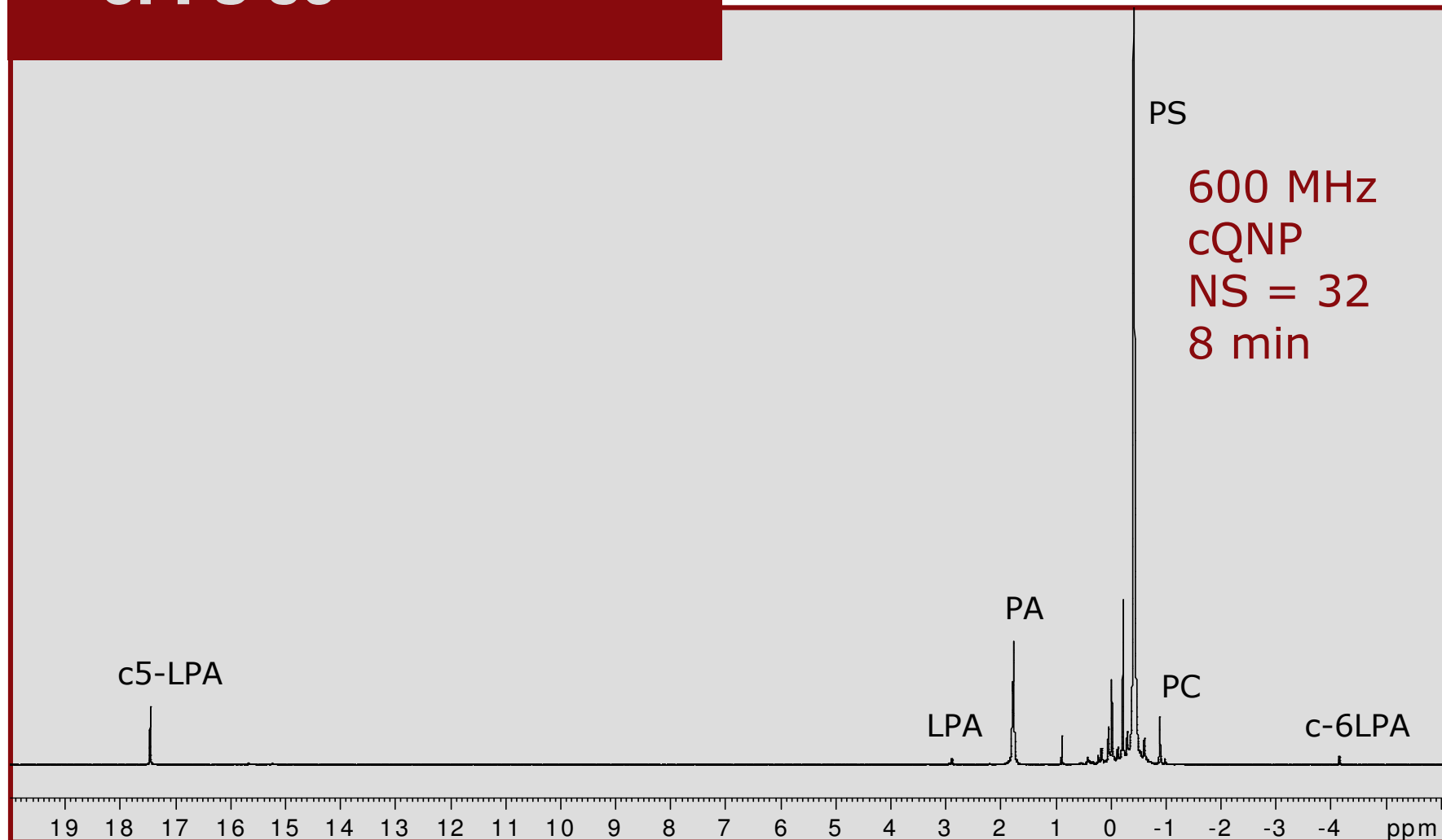


# **<sup>31</sup>P NMR Phospholipids of PS 50**

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# **<sup>31</sup>P NMR Phospholipids of PS 50**

Spectral Service

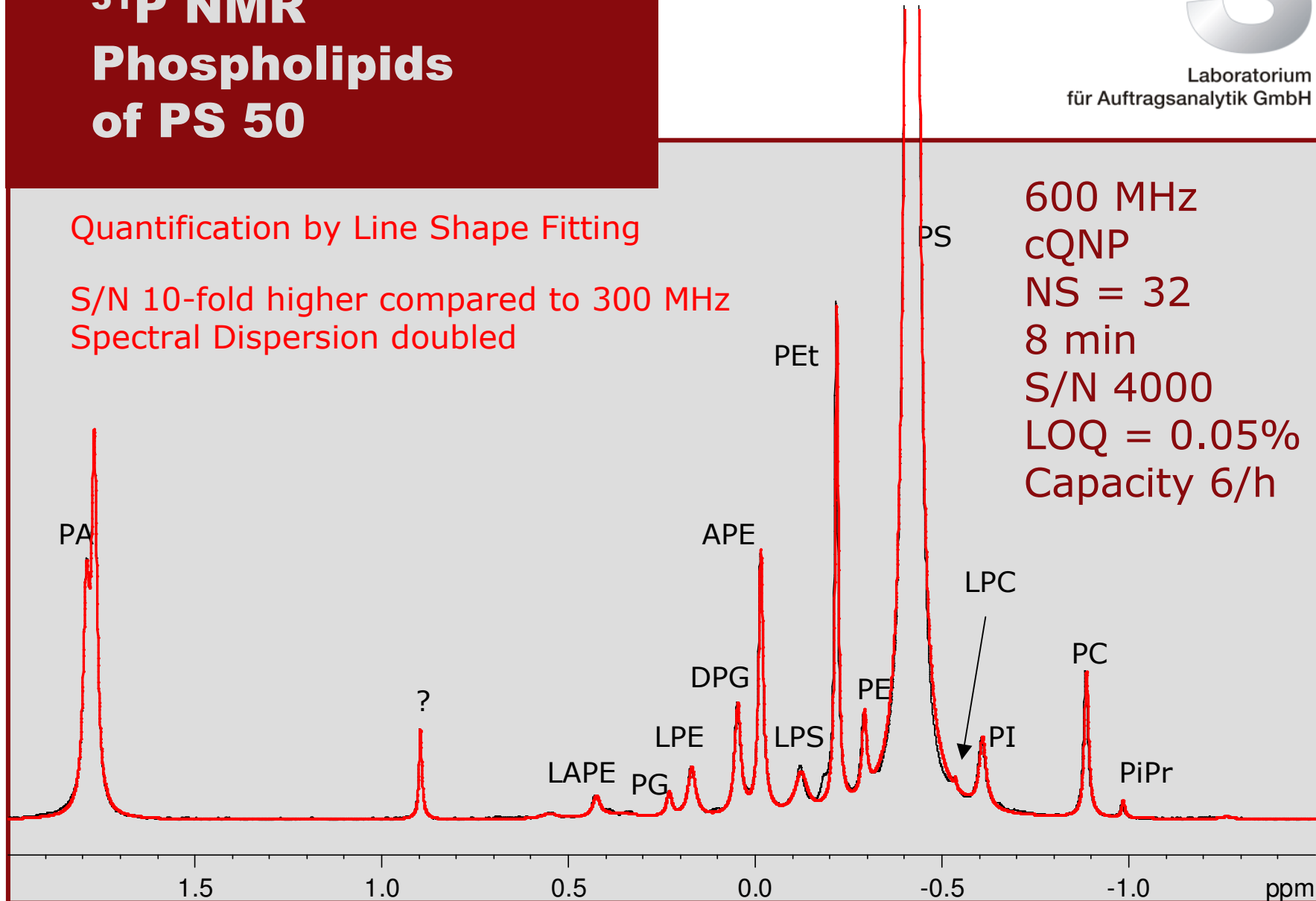


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Quantification by Line Shape Fitting

S/N 10-fold higher compared to 300 MHz  
Spectral Dispersion doubled

600 MHz  
cQNP  
NS = 32  
8 min  
S/N 4000  
LOQ = 0.05%  
Capacity 6/h



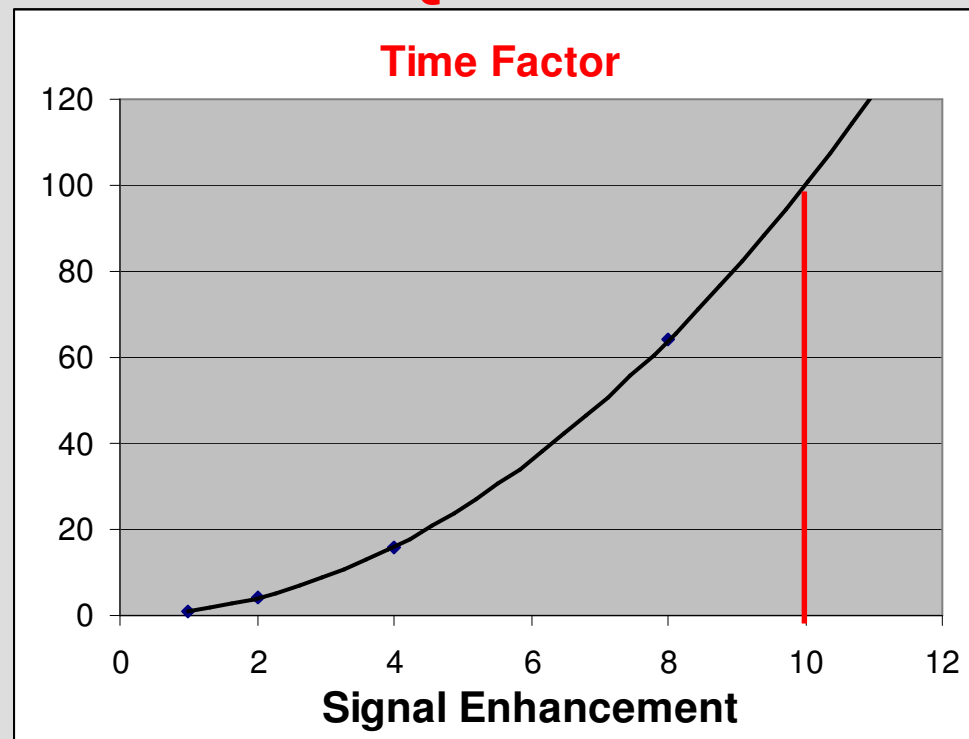


# Conclusion

and a little Promotion



## 600 MHz cQNP vs 300 MHz



8 min vs. 800 min

# Conclusion

and a little Promotion

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- Higher
- Faster
- Stronger

