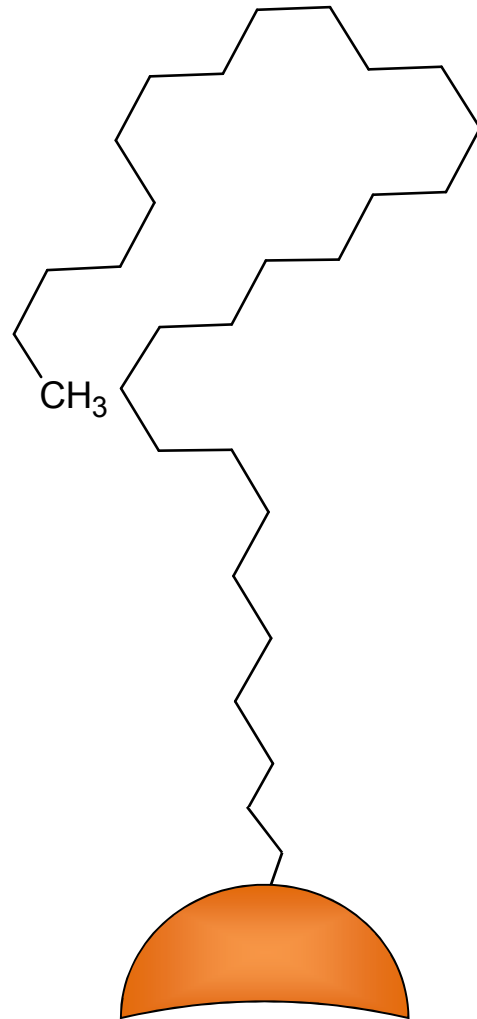


表面多孔性シリカを用いた C30固定相の評価

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(クロマニックテクノロジーズ)

C30カラムの注意点



トリアコンチル (C30)

結合密度が高いほど
異性体分離に優れる



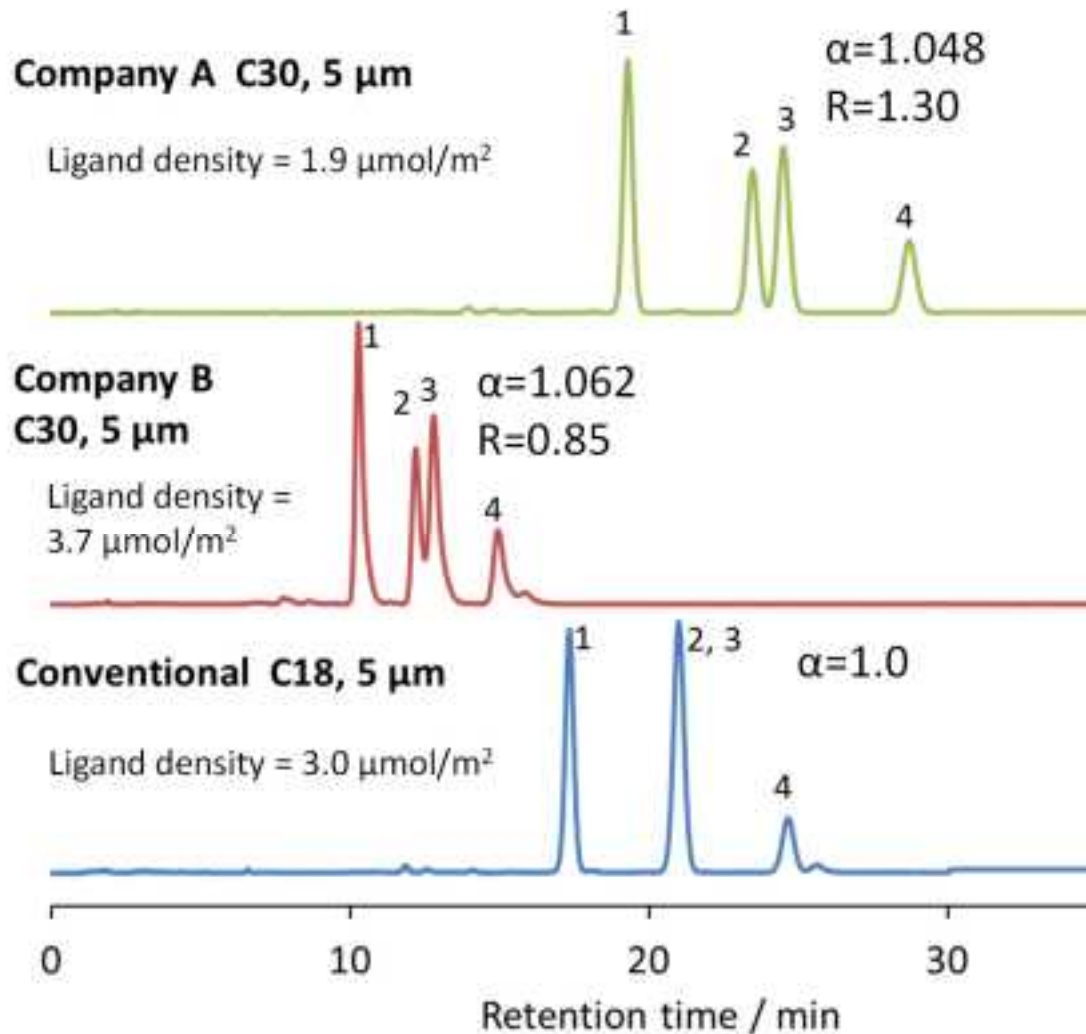
結合密度が高すぎると
ピークがテーリングする

結合密度が低いと
分離が不十分になる

結合密度の調整が重要

C30固定相の利点・問題点

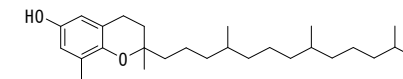
β/γ -tocopherolの分離係数・分離度



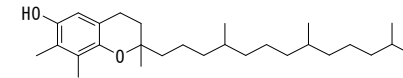
Column dimension: 250 x 4.6 mm
Mobile phase: Methanol/water = 97/3
Flow rate: 1.0 mL/min
Temperature: 30 °C
Detection: UV 295 nm

Sample,

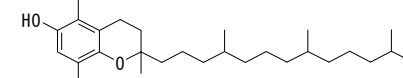
1 = δ -tocopherol



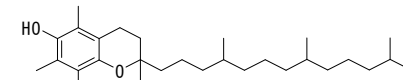
2 = γ -tocopherol



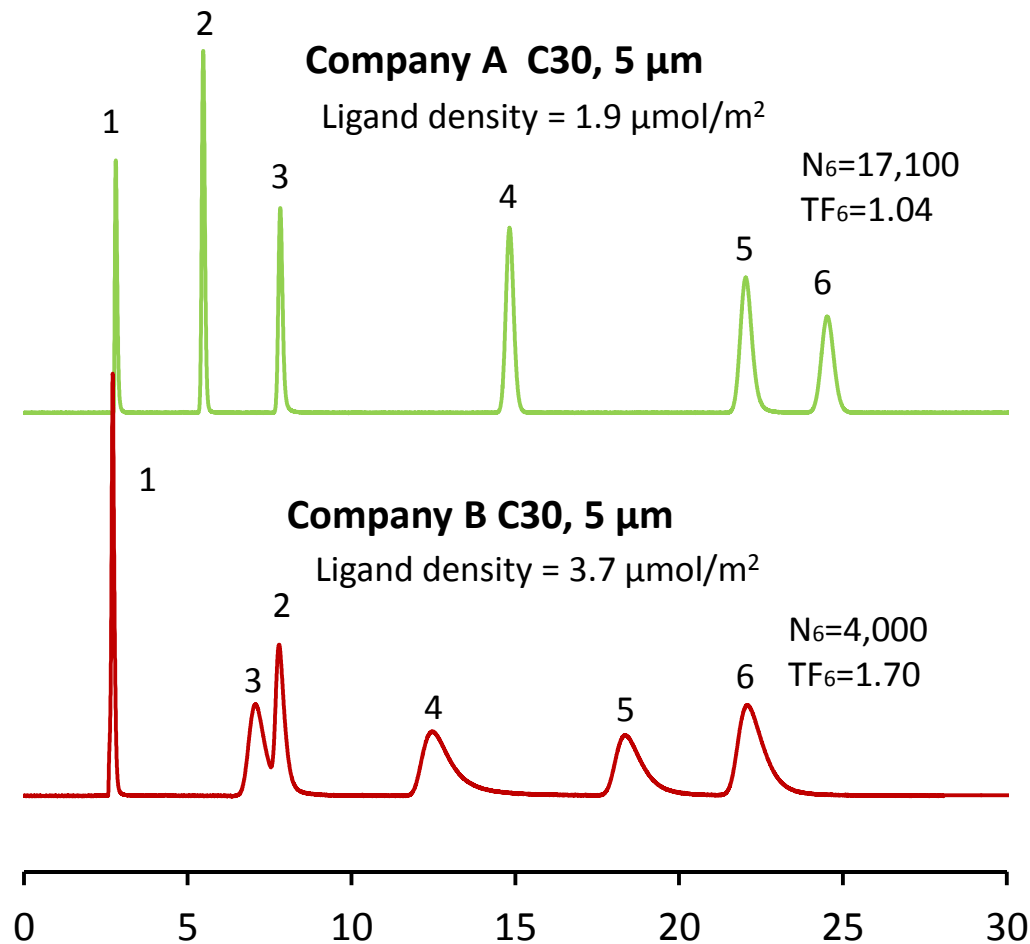
3 = β -tocopherol



4 = α -tocopherol



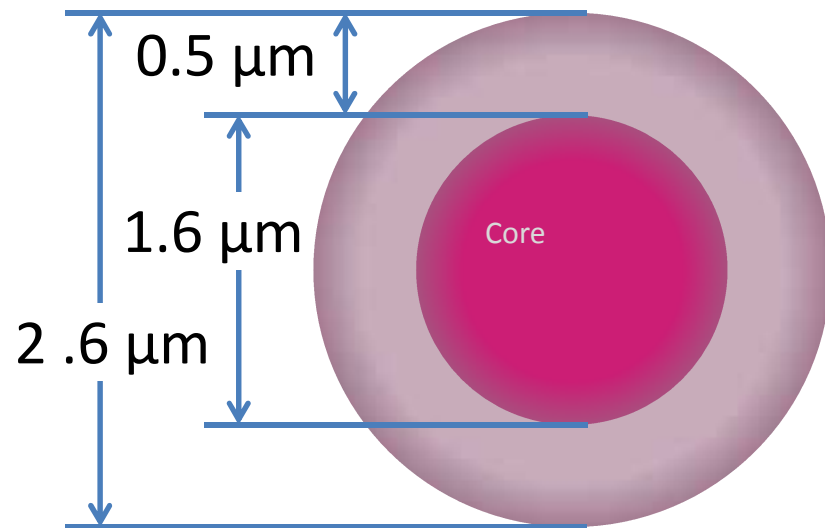
核酸塩基の分離



Column dimension, 250 x 4.6 mm
 Mobile phase: 10 mM phosphate buffer pH7.0
 Flow rate: 1.0 mL/min
 Temperature: 30 °C
 Detection: UV 254 nm
 Sample: 1 = Sodium Nitrite,
 2 = Cytosine
 3 = Uracil
 4 = Cytidine
 5 = Uridine
 6 = Thymine

移動相の有機溶媒の組成比に関わらず、
 結合密度の高いC30は段数が低くテーリングする。

使用した表面多孔質(コアシェル)シリカ



	細孔径	比表面積
1	10.2 nm	112 m ² /g
2	10.8 nm	106 m ² /g
3	12.0 nm	96.1 m ² /g
4	15.0 nm	79.6 m ² /g
5	15.5 nm	73.5 m ² /g

C30カラムの物性値と分離係数

Batch number		241215	241115	230216	110714	280314	220713	081112
Core shell silica	Particle diameter (μm)	2.6	2.6	2.6	2.6	2.6	2.6	2.6
	Thickness of porous layer (μm)	0.5	0.5	0.5	0.5	0.5	0.5	0.5
	Specific surface area ^a (m^2/g)	112	106	96.1	79.6	79.6	73.5	73.5
	Pore volume ^a (mL/g)	0.287	0.286	0.288	0.299	0.299	0.285	0.285
	Average pore diameter ^a (nm)	10.2	10.8	12.0	15.0	15.0	15.5	15.5
Carbon loading of only C30 (%)		6.59	5.48	6.08	4.43	5.40	4.10	5.20
Carbon loading including end-capping (%)		7.56	6.57	7.04	5.30	6.05	4.70	5.86
Ligand (C30) density ($\mu\text{mol}/\text{m}^2$)		1.79	1.55	1.91	1.64	2.03	1.64	2.11
Separation factor of β/γ -tocopherol ^b		1.0625	1.0570	1.0640	1.0376	1.0626	1.0415	1.0629
Volume of 1.5 nm thickness inside pore ^c ($\mu\text{L}/\text{m}^2$)		1.279	1.292	1.313	1.350	1.350	1.355	1.355
Ligand (C30) density ^d (mol/L)		1.400	1.200	1.455	1.215	1.504	1.210	1.557

a: Measured by Quantachrom Autosorb.

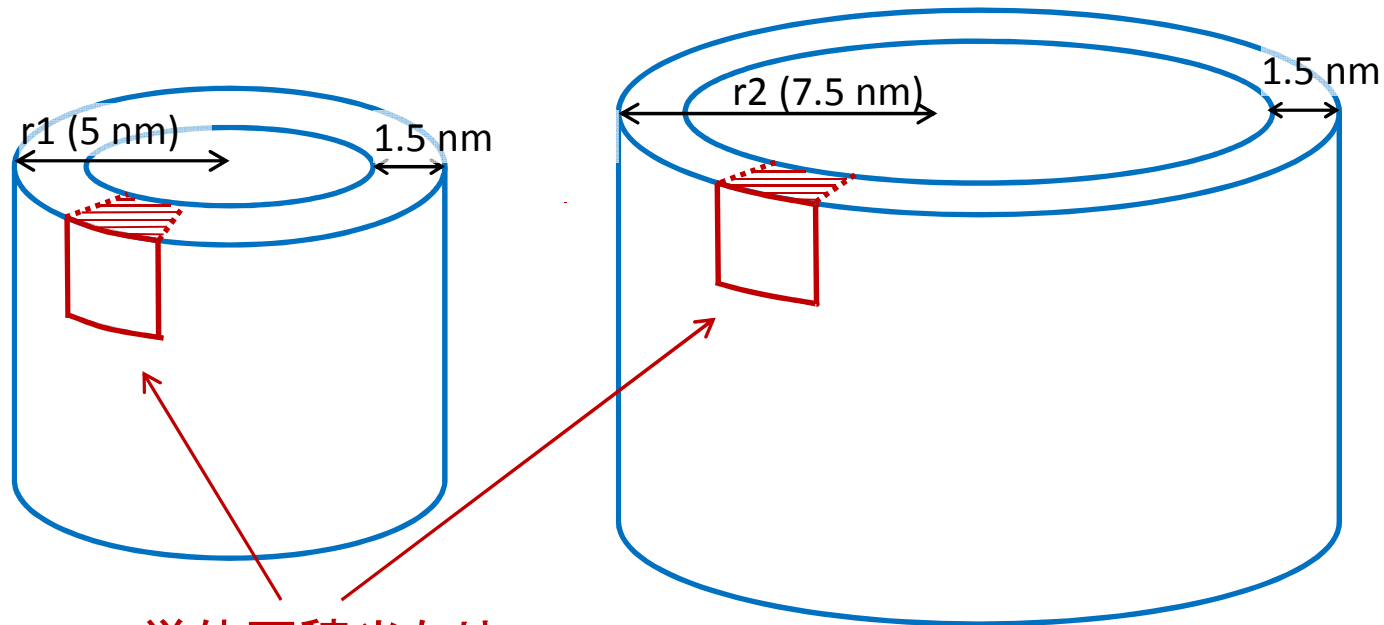
b: Mobile phase, methanol/water=97/3; temperature, 25 °C.

c: Postulated a pore as a cylinder with a same diameter, listed as the volume of per square meter.

d: Ligand density in the volume of 1.5 nm thickness inside of pore.

細孔表面から1.5nmの厚さの体積

Volume of 1.5 nm thickness inside pore ^d (L/m²)

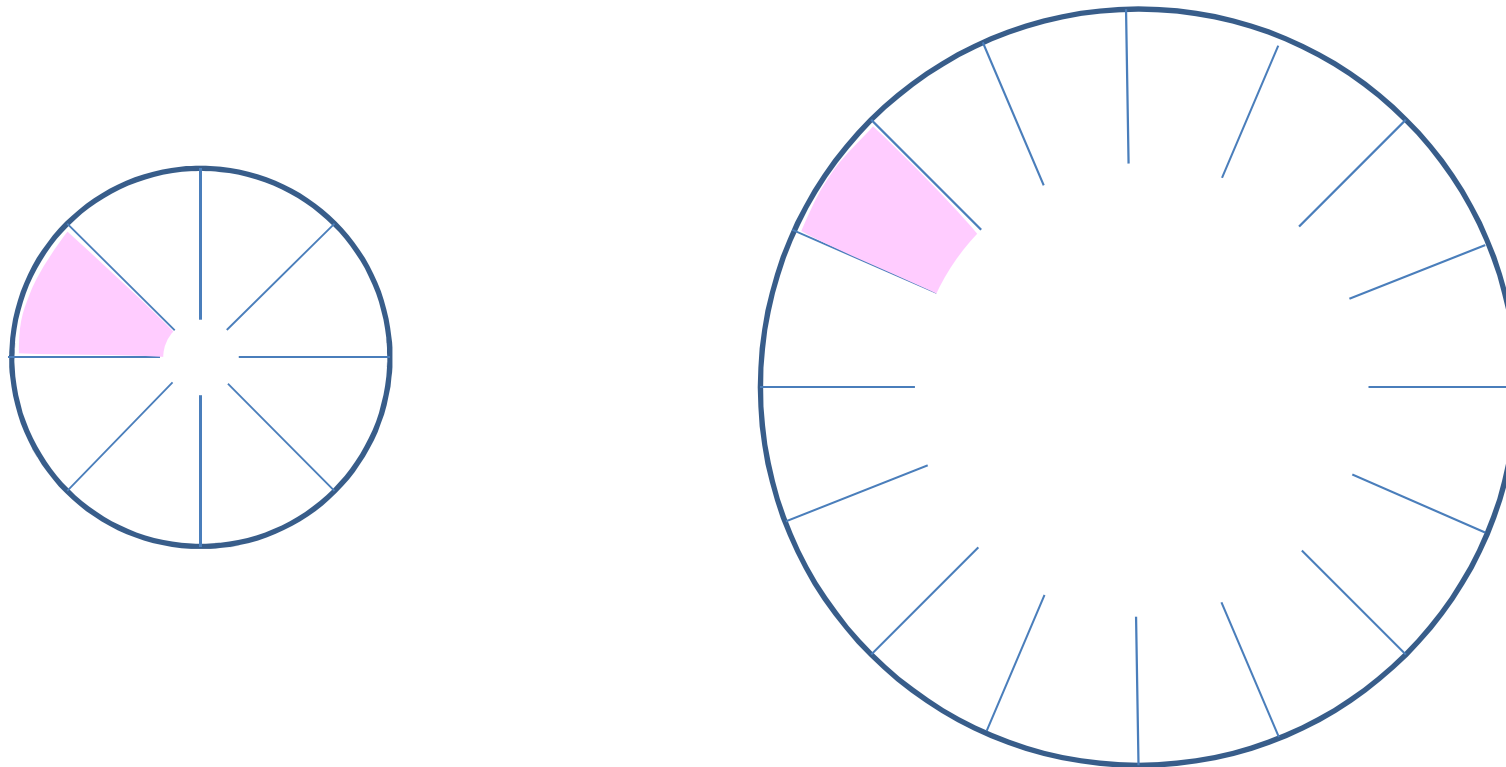


単位面積当たり

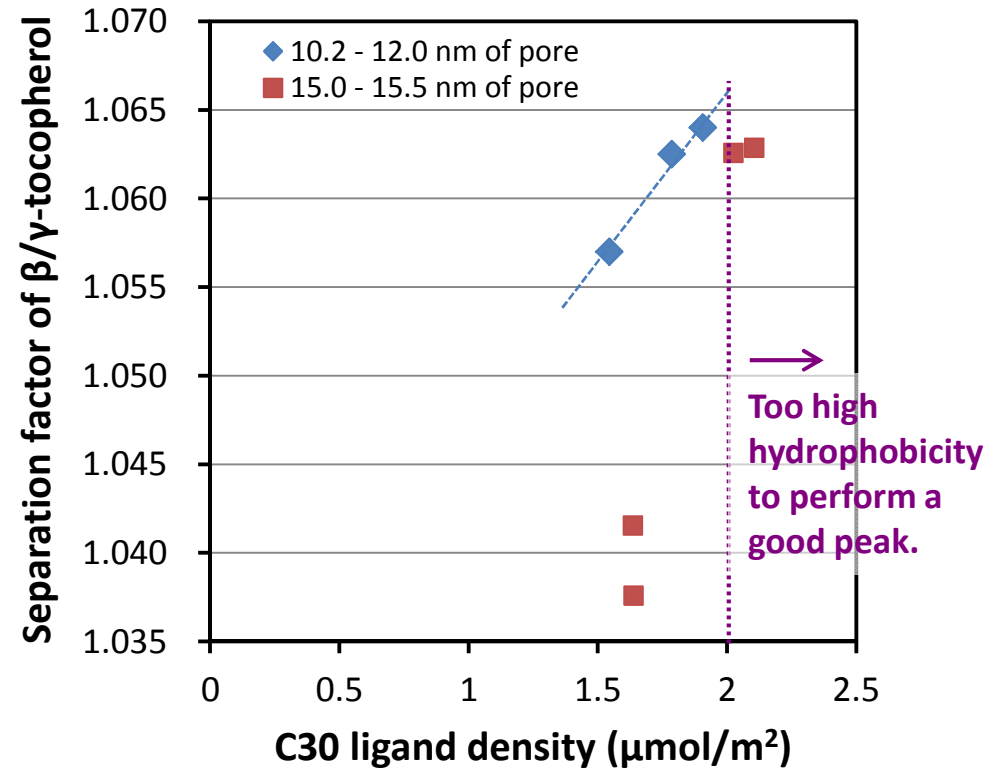
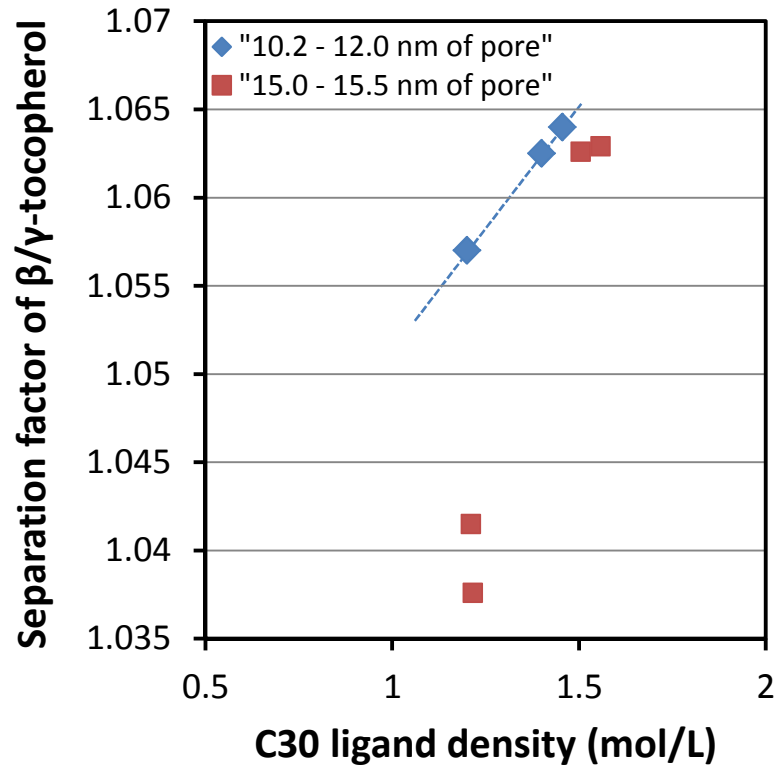
細孔を円柱と仮定・アルキル基は細孔表面から1.5nmの厚さに存在すると仮定



異なる細孔径での結合密度

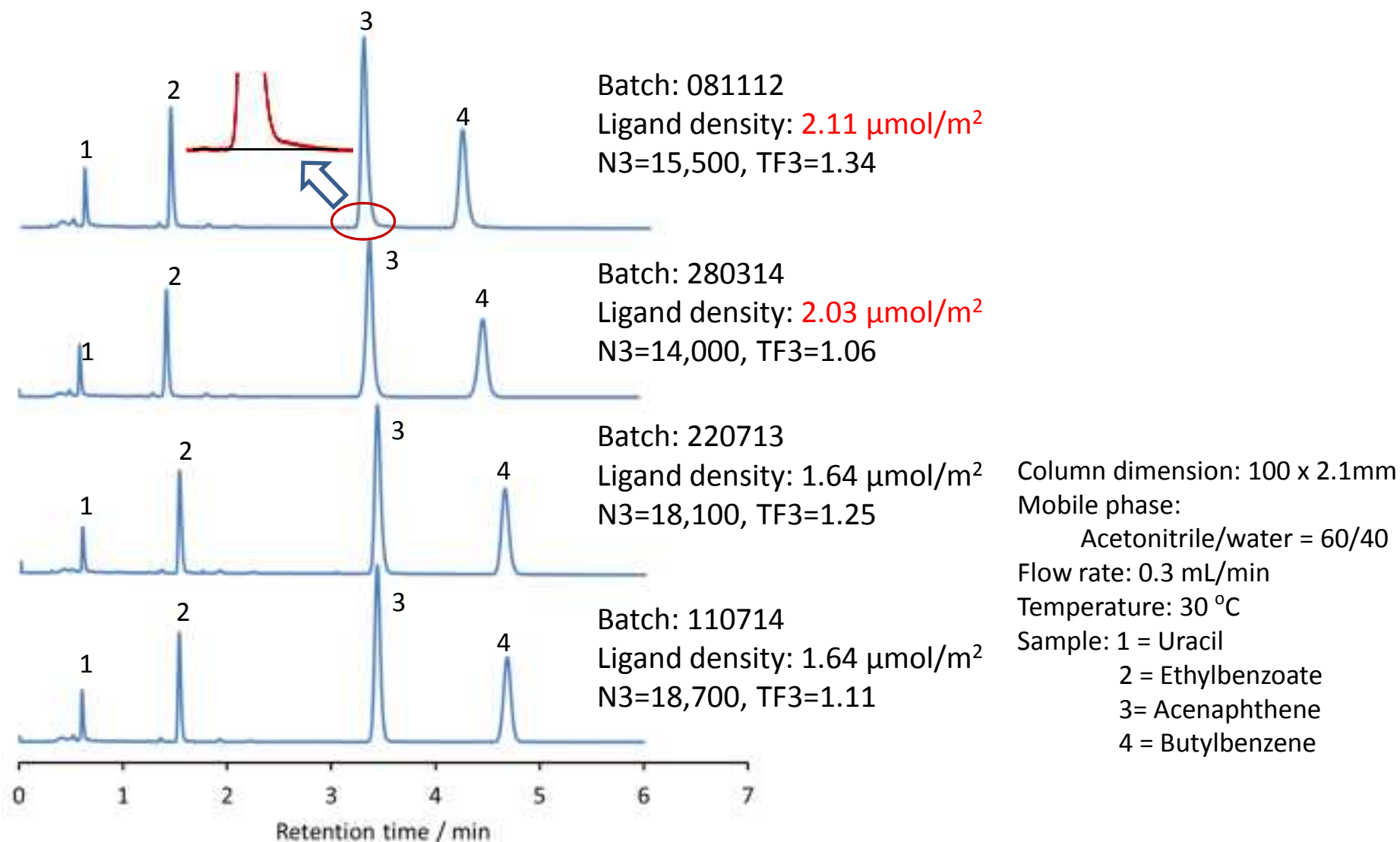


2種類の結合密度と分離係数

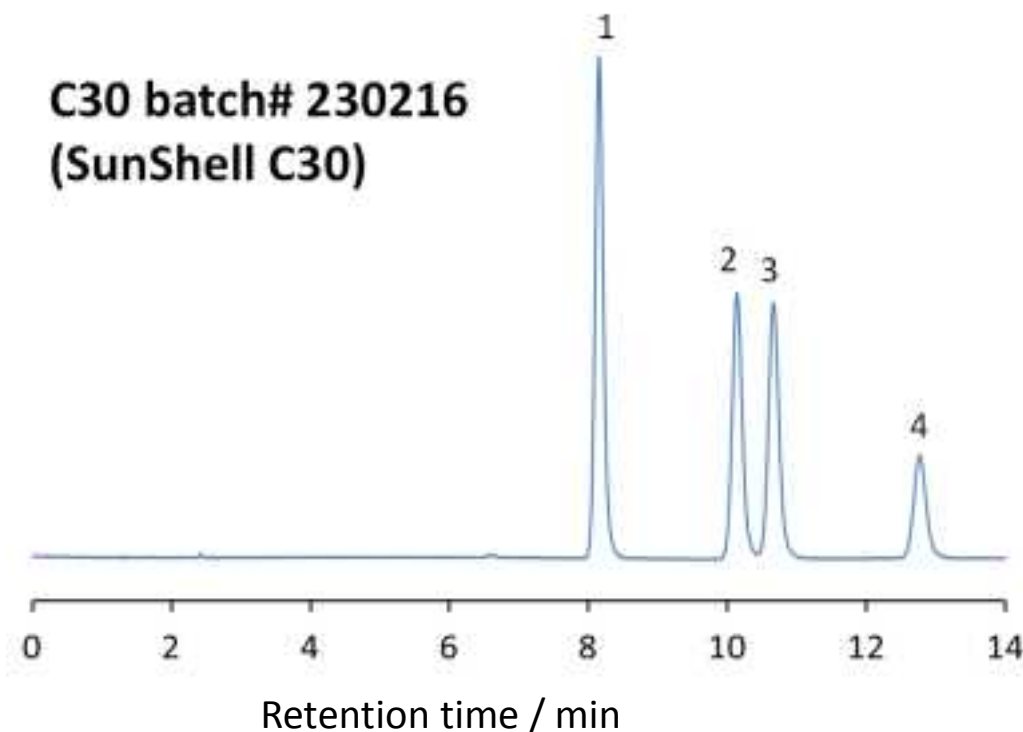


同じ結合密度でも細孔径によって分離係数が変化

結合密度と段数・テーリングファクターとの関係



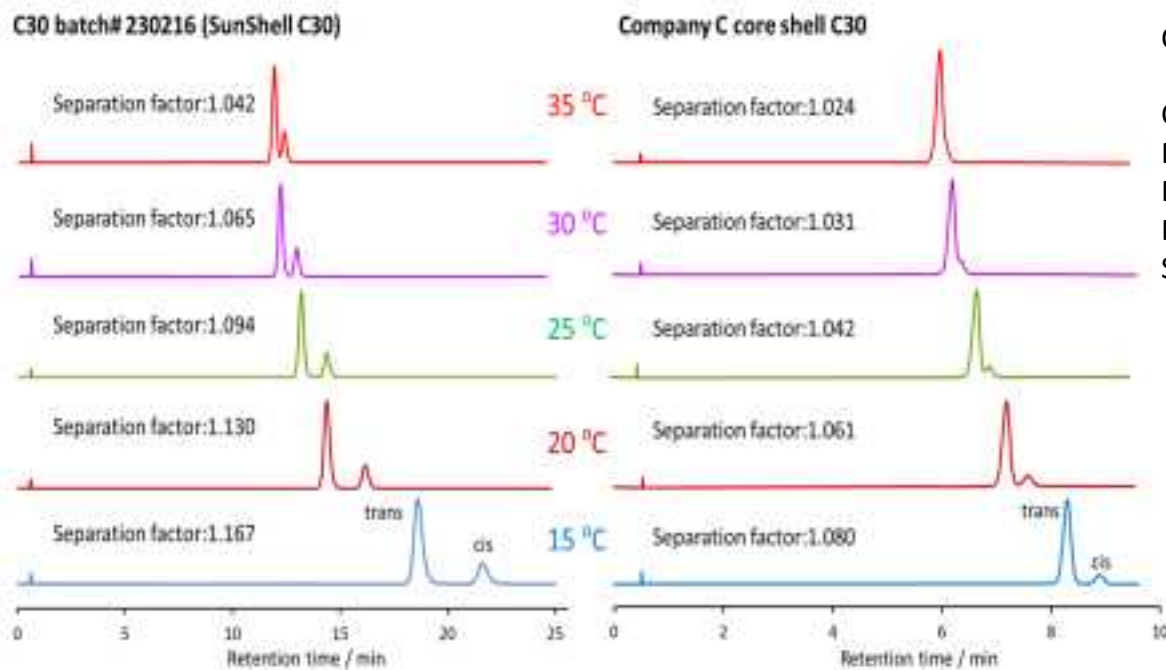
ビタミンE異性体の分離



Column: C30 batch# 230216, 2.6 μ m
150 x 3.0 mm
Mobile phase: Methanol/water = 97/3
Flow rate: 0.43 mL/min
Temperature: 25 $^{\circ}$ C
Detection: UV 295 nm
Sample: 1 = δ -tocopherol
2 = γ -tocopherol
3 = β -tocopherol
4 = α -tocopherol

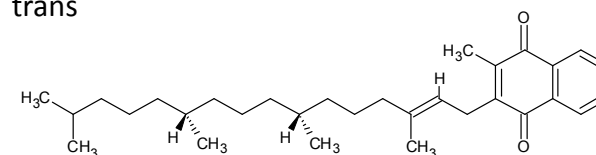
	Company A C30, 5 μ m 250 x 4.6 mm	Company B C30, 5 μ m 250 x 4.6 mm	SunShell C30, 2.6 μ m 150 x 3.0 mm
Separation factor of β/γ -tocopherol	1.048	1.062	1.064
Resolution of β/γ -tocopherol	1.30	0.85	2.01

ビタミンK1の異性体の分離

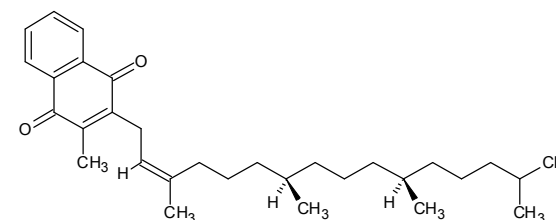


Column: C30 batch# 230216 (SunShell C30), 2.6 μm
 Company C core shell C30, 2.6 μm
 Column size: 100 x 2.1 mm
 Mobile phase: methanol/water = 96/4
 Flow rate: 0.35 mL/min;
 Detection: UV 250 nm;
 Sample: vitamin K1 isomers (trans and cis).

trans



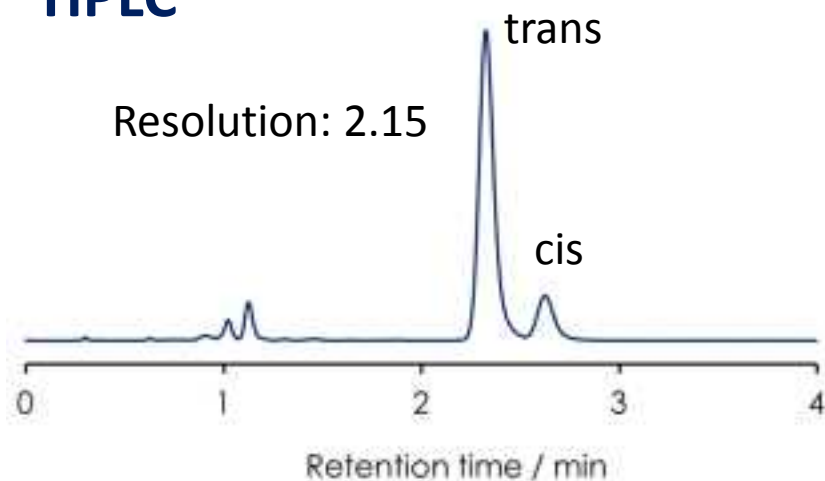
cis



	Vitamin K1 isomers (trans and cis)			
	Separation factor		Resolution	
	C30 batch# 230216	Company C C30	C30 batch# 230216	Company C C30
35 °C	1.042	1.024	1.23	---
30 °C	1.065	1.031	1.94	---
25 °C	1.094	1.042	2.88	0.91
20 °C	1.130	1.061	3.33	1.41
15 °C	1.167	1.080	3.79	1.78

ビタミンK1の異性体の高速分離

HPLC



HPLC condition

Column: SunShell C30, 2.6 μm 50 x 3.0 mm

Mobile phase: Methanol

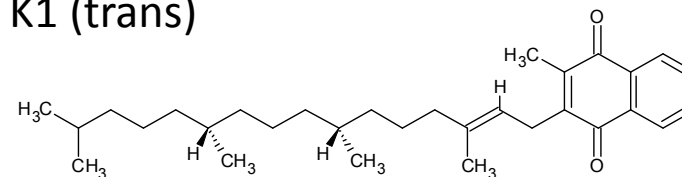
Flow rate: 0.80 mL/min

Temperature: 15 $^{\circ}\text{C}$

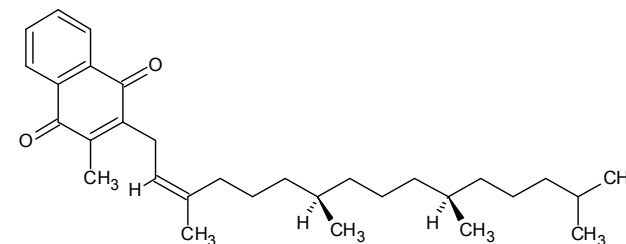
Detection: UV 250 nm

Sample, vitamin K1 isomers (trans and cis).

Vitamin K1 (trans)



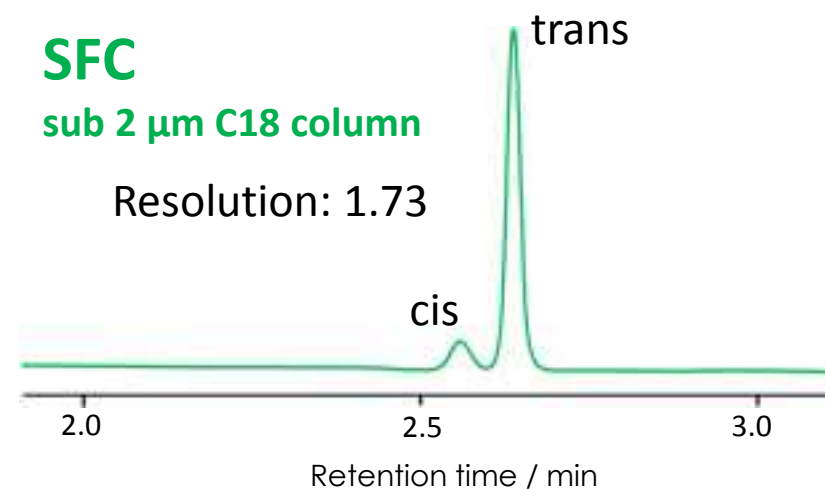
Vitamin K1 (cis)



SFC

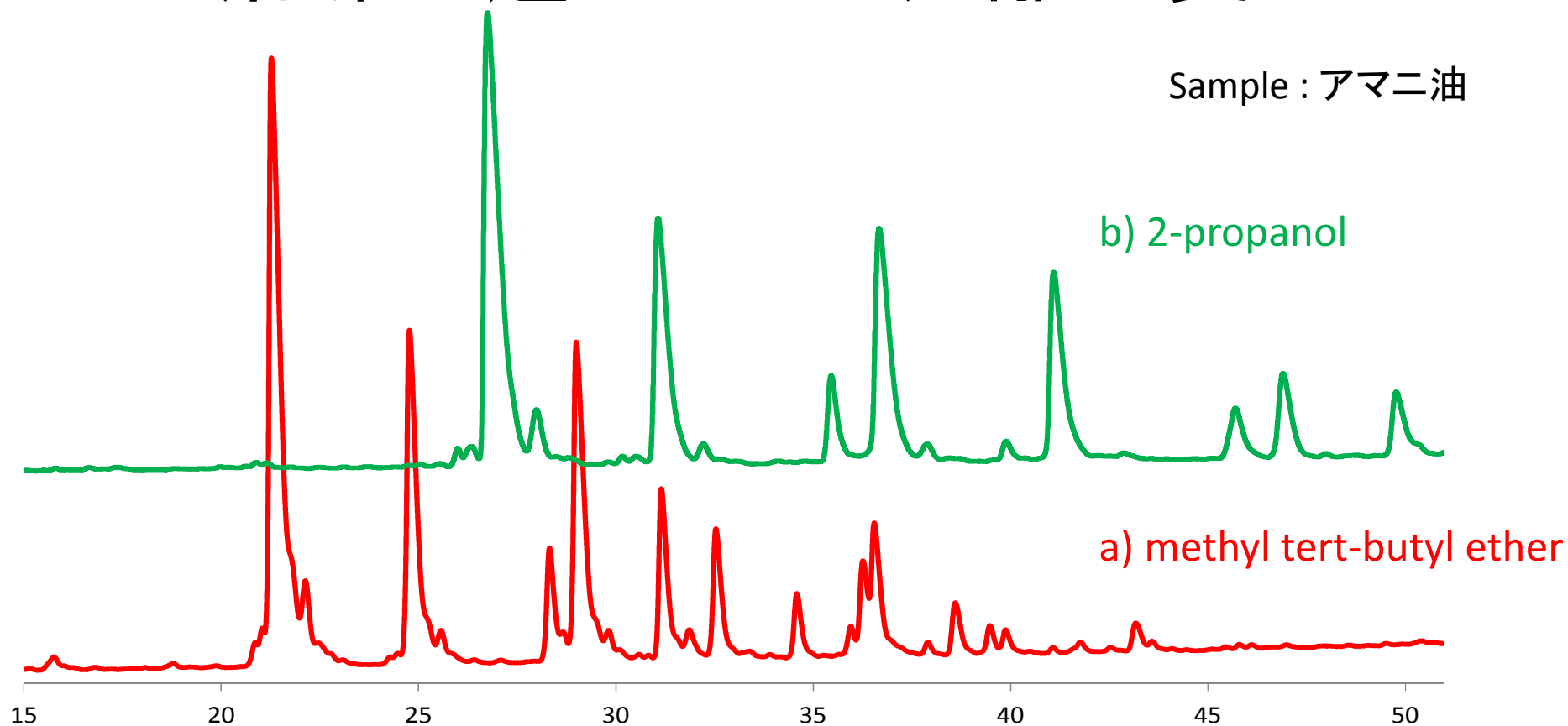
sub 2 μm C18 column

Resolution: 1.73



溶媒の違いによる分離の変化

Sample : アマニ油



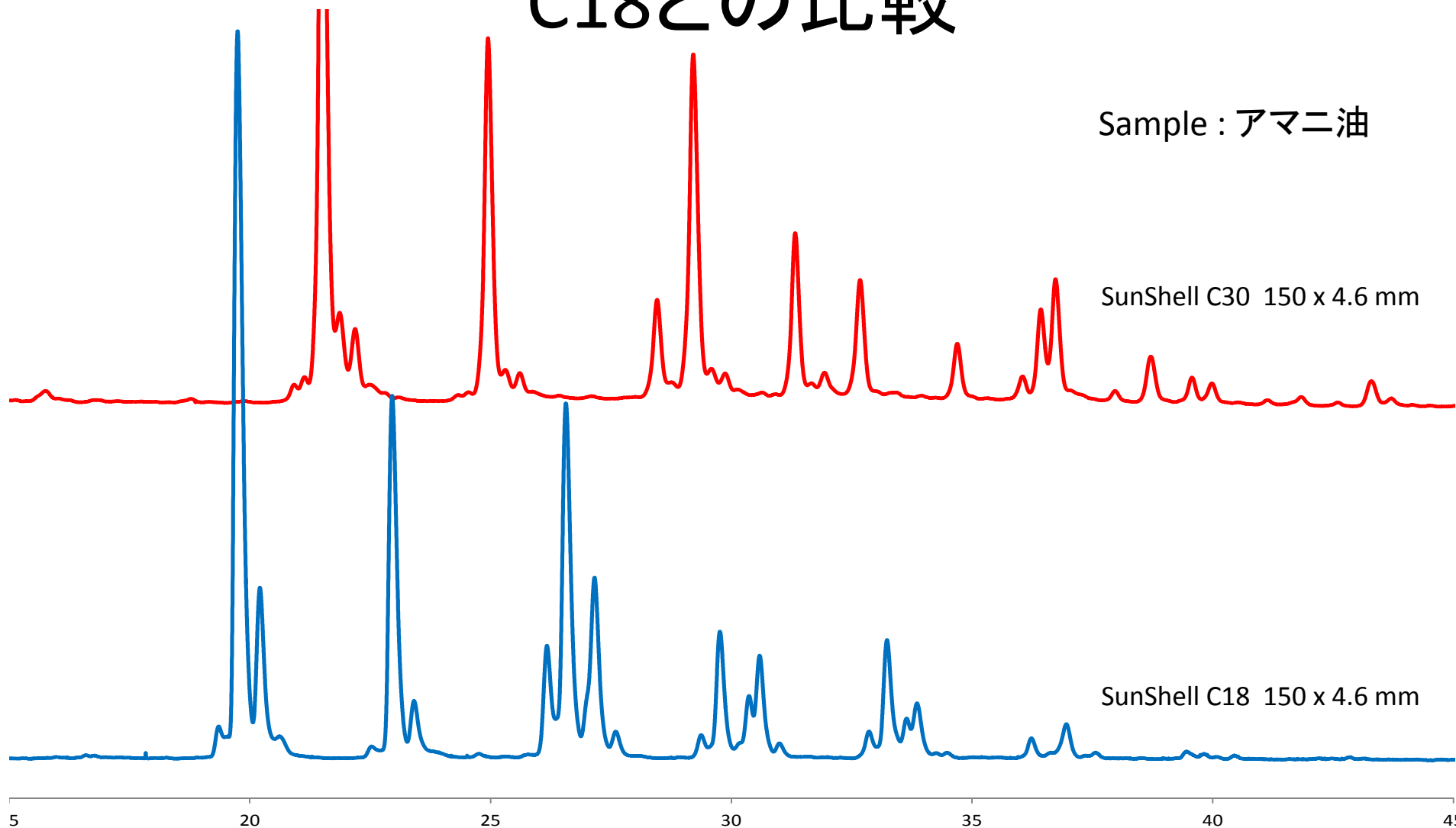
Column dimension, 150 x 4.6 mm; flow rate, 1.0 mL/min; temperature, 25 °C
 mobile phase : A:acetonitrile,, B: methyl tert-butyl ether (MTBE) B% 5 to 50 in 55 min
 mobile phase : A:acetonitrile,, B:2-propanol B% 10 to 50 in 55 min

C18との比較

Sample : アマニ油

SunShell C30 150 x 4.6 mm

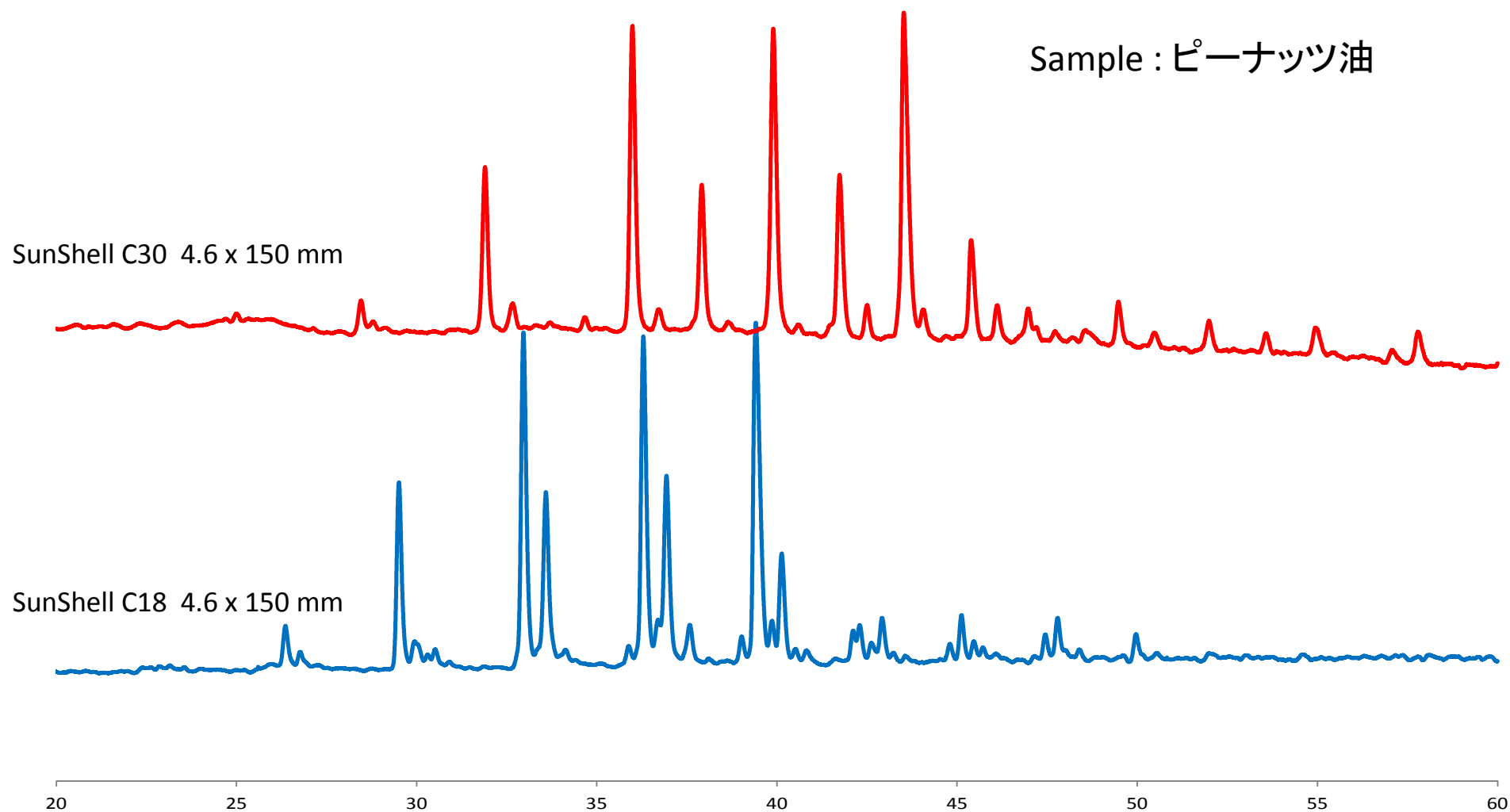
SunShell C18 150 x 4.6 mm



Column dimension, 150 x 4.6 mm; flow rate, 1.0 mL/min; temperature, 25 °C
mobile phase : A:acetonitrile,, B:MTBE B% 10 to 50 in 55 min

C18との比較2

Sample : ピーナッツ油



Column dimension, 150 x 4.6 mm; flow rate, 1.0 mL/min; temperature, 25 °C
mobile phase : A:acetonitrile,, B:MTBE B% 10 to 50 in 55 min

まとめ

1. 長鎖アルキル基は脂溶性化合物(ビタミンE, K1)の異性体分離に優れている。
2. 長鎖アルキル基の結合密度が高い方が異性体の分離は向上するが, 結合密度が $2 \mu\text{mol}/\text{m}^2$ 以上になると段数が低下し, テーリング傾向になる。
3. 細孔径により, 単位面積当たりの結合密度($\mu\text{mol}/\text{m}^2$)が同じでも, 単位体積当たりの結合密度(mol/L)は変化する。
4. 単位面積当たりの結合密度が高すぎず, 単位体積当たりの結合密度の高い充填剤が, 段数, テーリング等の影響を受ける分離度に対して, 最も高い値を示した。