

Hydrophilic Interaction Chromatography

ULTRON AF-HILIC-CD Series



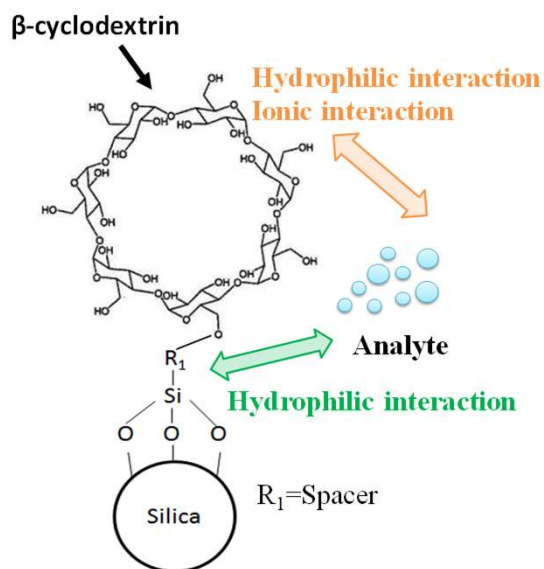
SHINWA CHEMICAL INDUSTRIES LTD.

About ULTRON AF-HILIC-CD series

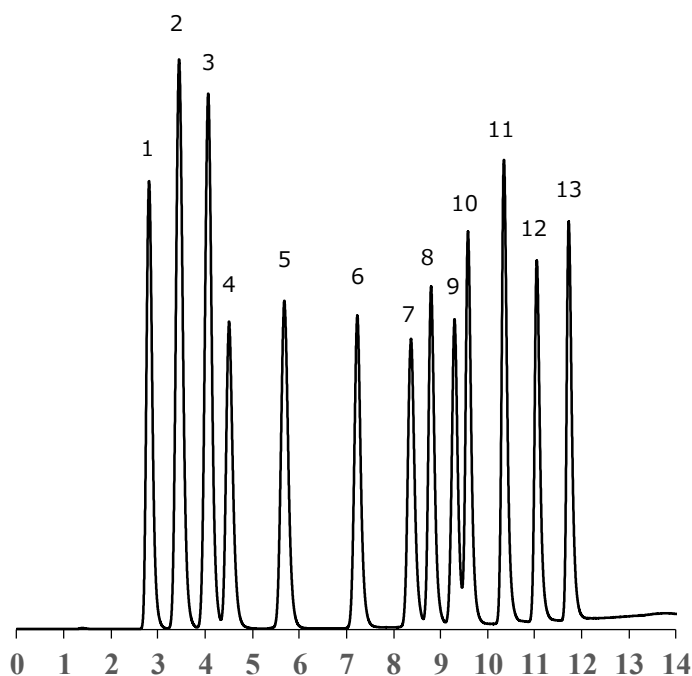
ULTRON AF-HILIC-CD is a HILIC mode column in which β -cyclodextrin (⊗) is chemically bonded to a silica stationary phase via a spacer. The selectivity towards molecular structure is high, and also shows high resolution analysis for structural analogues and structural isomers of acidic, basic and neutral compounds.

(⊗) CD has a hollow cavity derived from its cyclic structure and are hydrophobic on the inside and hydrophilic on the outside. It is characterized by the inclusion complex formation that takes with various compounds by the intermolecular force inside the cyclic structure.

Hydrophilic Interaction Chromatography (HILIC) is a separation mode used for analysis of polar compounds that are difficult to retain in reversed phase column. Polar compounds elute later than hydrophobic compound. In case of analysis of hydrophilic compounds, ion pair reagents are often used in order to retain in reversed phase columns, but in HILIC it is possible to retain and separate without using this ion pair reagent.



Simultaneous analysis of mono-, di- and oligosaccharides



- | | |
|--------------|--------------------|
| 1. Xylose | 8. Lactose |
| 2. Fructose | 9. Trehalose |
| 3. Glucose | 10. Xylotriose |
| 4. Mannitol | 11. 1-Kestose(GF2) |
| 5. Xylobiose | 12. Raffinose |
| 6. Sucrose | 13. Nystose(GF3) |
| 7. Lactulose | |

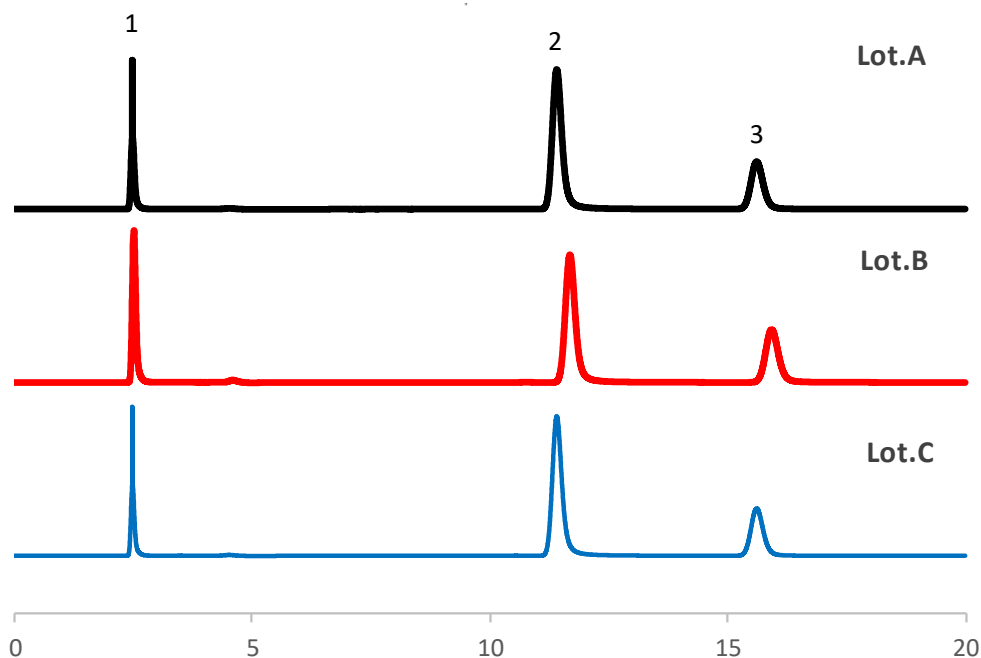
Gradient : 10 minutes gradient, 88% - 65% with 20minutes re-equilibration

Time (min)	%A	%B
0.00	96	4
10.00	58	42
10.01	96	4
30.00	96	4

Column: ULTRON AF-HILIC-CD (HT) (2 μ m)
 Column size: 100 mm x 2.0 mm I.D.
 Mobile phase: A 20 mM CH₃CO₂NH₄ (pH 6.8) /CH₃CN = 10/90 (v/v)
 B 20 mM CH₃CO₂NH₄ (pH 6.8) /CH₃CN = 70/30 (v/v)

Flow rate: 0.26 mL/min
 Column temp.: 40°C
 Detection: ELSD
 Injection vol.: 2.5 μ L

ULTRON AF-HILIC-CD series :Reproducibility

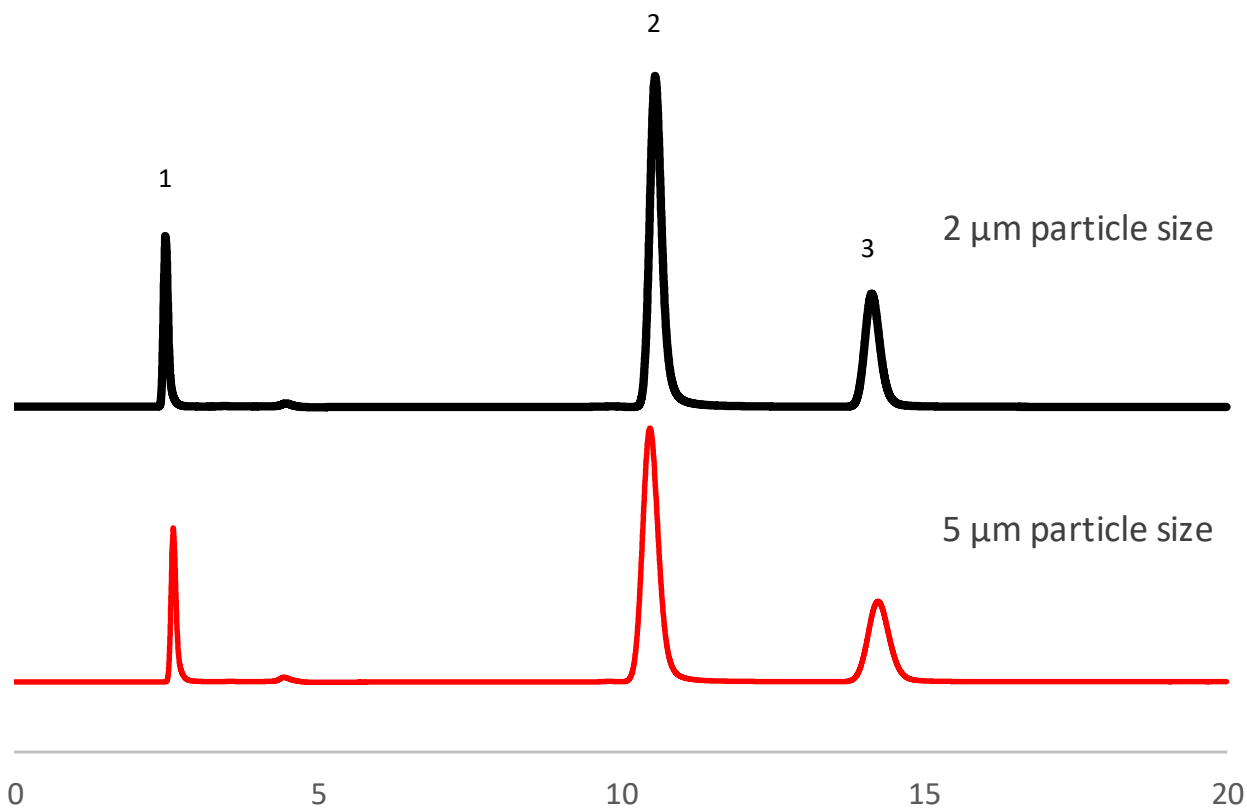


Column: ULTRON AF-HILIC-CD (HT) (2 μ m)
 Column size: 150 mm L x 2.0 mm I.D.
 Mobile phase: 20 mM CH₃CO₂NH₄ (pH 4.7)/CH₃CN = 10/90 (v/v)
 Samples: 1. Toluene 2. Adenosine 3. Vidarabine

Flow rate: 0.113 mL/min
 Column temp.: 30°C
 Detection: UV-254 nm
 Injection vol.: 0.5 μ L

Packing material Lot.	Rt. Peak 2	Rt. Peak 3	Separation factor
Lot. A	11.4	15.61	1.47
Lot. B	11.68	15.92	1.46
Lot. C	11.41	15.62	1.47
Average	11.5	15.72	1.47
RSD(%)	1.12	0.92	0.29

ULTRON AF-HILIC-CD series: Scalability (2 μm , 5 μm)



Column: ULTRON AF-HILIC-CD (HT) (2 μm)/ ULTRON AF-HILIC-CD (5 μm)
 Column size: 100 mm x 2.0 mm I.D./100 mm x 2.0 mm I.D.
 Flow rate: 0.10 mL/min
 Mobile phase: 20 mM $\text{CH}_3\text{CO}_2\text{NH}_4$ (pH 4.7)/ CH_3CN = 10/90 (v/v)
 Samples: 1. Toluene 2. Adenosine 3. Vidarabine
 Column temp.: 30°C
 Detection: UV-254 nm
 Injection vol.: 0.8 μL

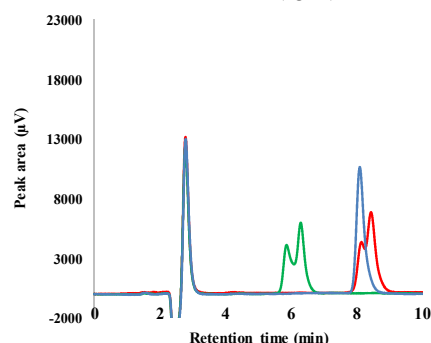
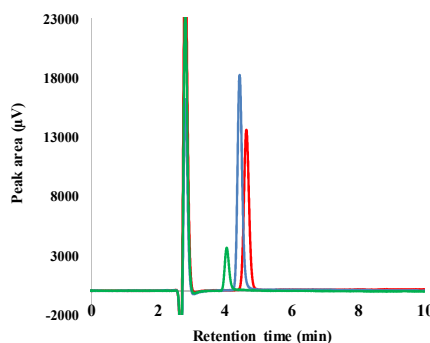
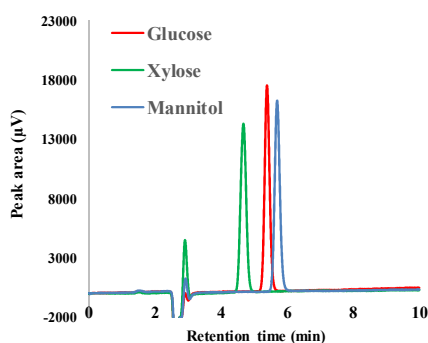
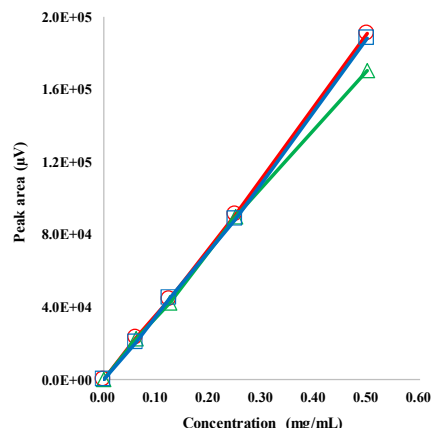
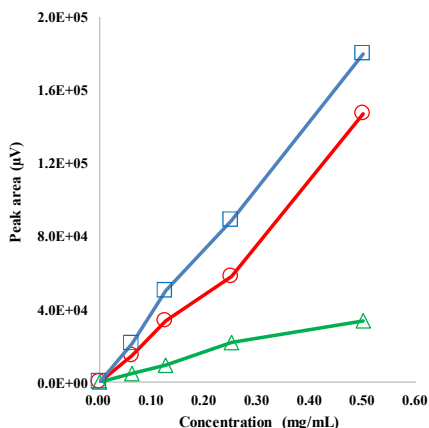
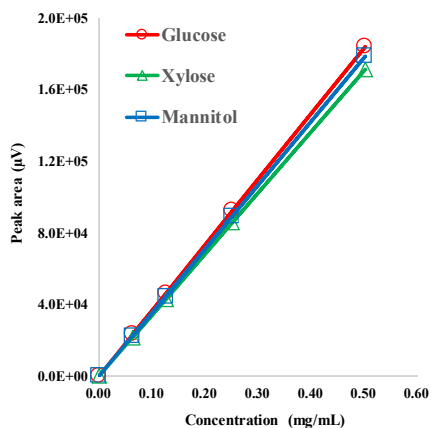
Particle size	Rt. Peak 2	Rt. Peak 3	Separation factor
2 μm	10.57	14.14	1.44
5 μm	10.49	14.25	1.47

Comparison of Schiff base formation and anomer separation

ULTRON AF-HILIC-CD

Amine column

Amide column



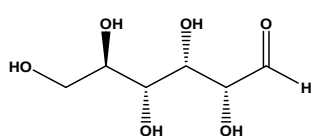
Schiff base formation: No
Anomer separation: No

Schiff base formation: Yes
Anomer separation: No

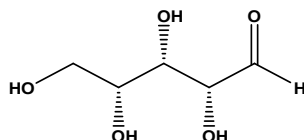
Schiff base formation: Yes
Anomer separation: Yes

Column: ULTRON AF-HILIC-CD
Column size: 100 mm x 4.6 mm I.D.
Mobile phase: H₂O /CH₃CN = 25/75 (v/v)

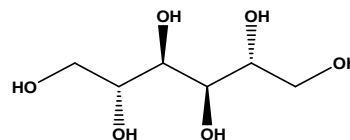
Flow rate: 0.5 mL/min
Column temp.: 30°C
Detection: RID
Injection vol.: 20 µL



Glucose
(reducing sugar)

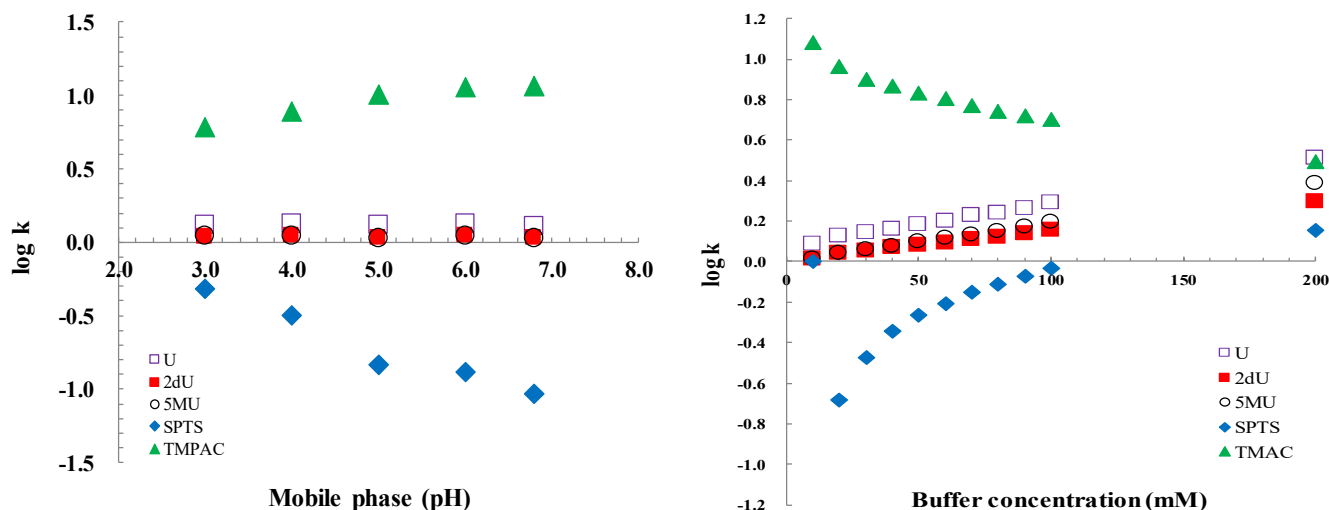


Xylose
(reducing sugar)



Mannitol
(non-reducing sugar alcohol)

Effect of mobile phase pH and buffer concentration on retention behavior



Column: ULTRON AF-HILIC-CD (HT) (2 μ m) Flow rate: 0.1 mL/min
 Column size: 100 mm L x 2.0 mm I.D. Column temp.: 25°C
 Mobile phase: 20 mM CH₃CO₂NH₄ (pH XX)/CH₃CN = 10/90 (v/v) Detection: UV-254 nm
 YY mM CH₃CO₂NH₄ (pH 4.7)/CH₃CN = 10/90 (v/v) Injection vol.: 0.8 μ L
 Samples: Uridine (U), 2'-Deoxyuridine (2dU), 5-Methyluridine (5MU), Toluene (Tol),
 Sodium p-toluenesulfonate (SPTS), N,N,N-trimethylphenyl ammonium chloride (TMPAC)

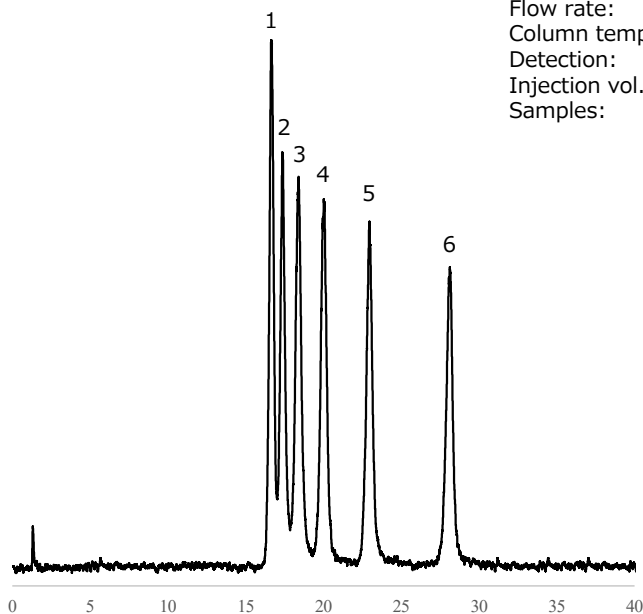
Effect of mobile phase pH

- U, 2dU, 5MU: Regardless of mobile phase buffer pH, the retention tendency was constant.
- SPTS: The retention time tends to decrease with the increase of mobile phase buffer pH value.
- TMPAC: The retention time tends to increase with the increase of mobile phase buffer pH value.

Effect of buffer concentration

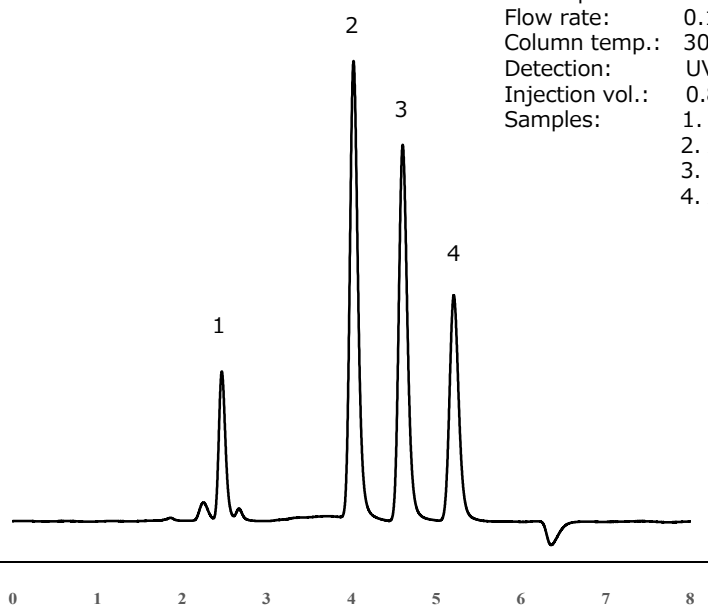
- U, 2dU, 5MU: The retention time tends to increase gradually with the increase of buffer concentration.
- SPTS/TMPAC: Rapid increase/decrease in retention time between 10 and 50 mmol/L.

Separation of sugar isomers



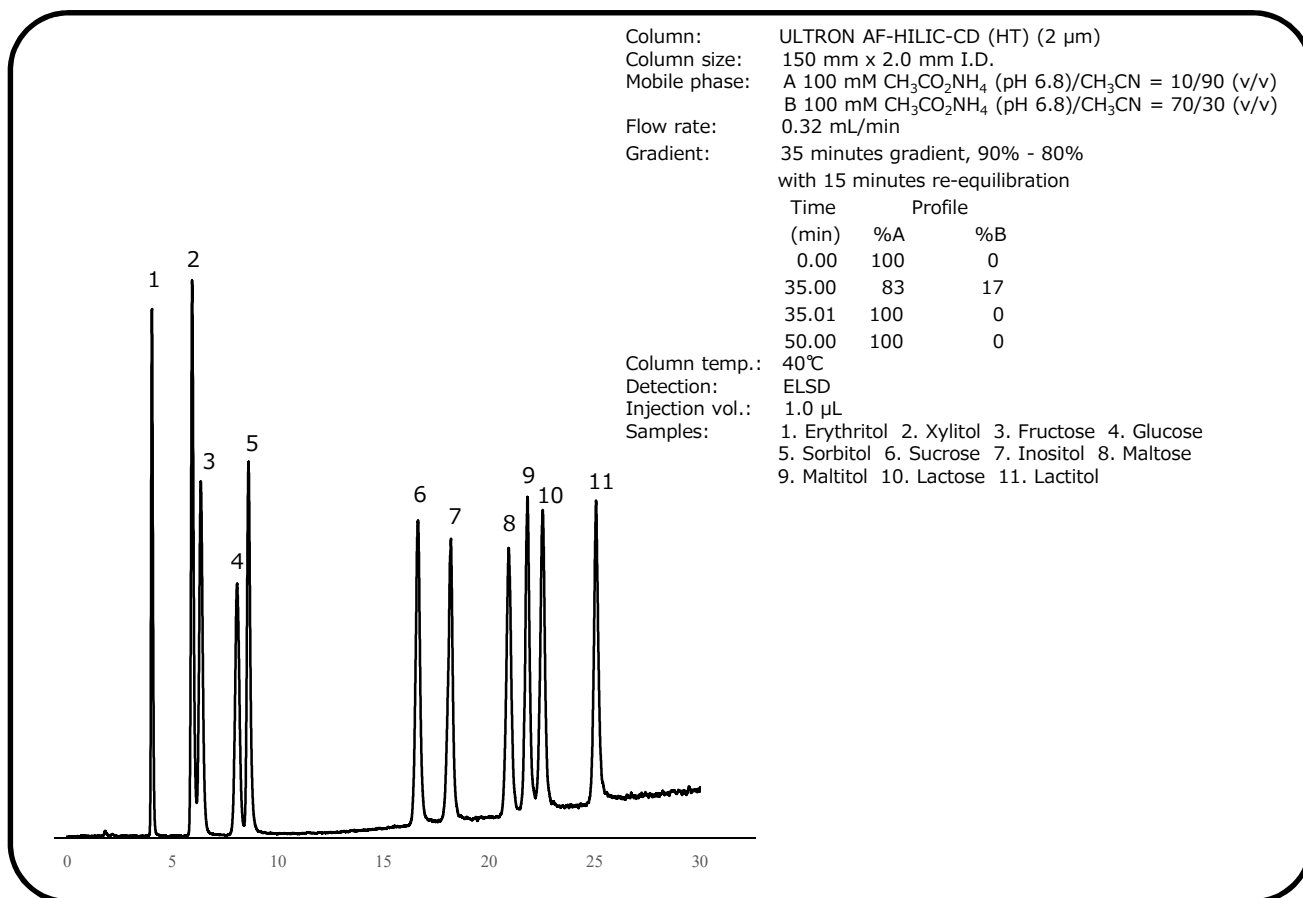
Column: ULTRON AF-HILIC-CD (HT) (2 μ m)
Column size: 150 mm x 2.0 mm I.D.
Mobile phase: 100 mM $\text{CH}_3\text{CO}_2\text{NH}_4$ (pH 6.8)/ CH_3CN = 10/90 (v/v)
Flow rate: 0.45 mL/min
Column temp.: 40°C
Detection: ELSD
Injection vol.: 1.0 μ L
Samples:
1. Turanose (Glucose - Fructose $\alpha(1 \rightarrow 3)$)
2. Palatinose (Glucose - Fructose $\alpha(1 \rightarrow 6)$)
3. Maltulose (Glucose - Fructose $\alpha(1 \rightarrow 4)$)
4. Maltose (Glucose - Glucose $\alpha(1 \rightarrow 4)$)
5. Lactose (Galactose - Glucose $\beta(1 \rightarrow 4)$)
6. Trehalose (Glucose - Glucose $\alpha(1 \rightarrow 1)$)

High-throughput analysis of melamine related compounds



Column: ULTRON AF-HILIC-CD (HT) (2 μ m)
Column size: 100 mm x 2.0 mm I.D.
Mobile phase: 20 mM HCO_2NH_4 (pH 6.8)/ CH_3CN = 18/82 (v/v)
Flow rate: 0.15 mL/min
Column temp.: 30°C
Detection: UV-220 nm
Injection vol.: 0.8 μ L
Samples:
1. Cyanuric acid
2. Ammelide
3. Melamine
4. Ammeline

Simultaneous analysis of sugars and sweeteners



ULTRON AF-HILIC-CD Series line-up

Product name	Particle (μ m)	Column size (mm)
ULTRON AF-HILIC-CD (HT)	2	50 x 2.0
		100 x 2.0
		150 x 2.0
ULTRON AF-HILIC-CD	5	100 x 2.0
		100 x 4.6
		150 x 2.0
		150 x 4.6
		250 x 4.6

※ Please inquire about other column sizes

Please feel free to contact us with questions related to analyses. Column screening services are also available. Please be aware that specifications and prices are subject to change without prior notification.

Contact details for inquiries

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 Kyoto 612-8307 Japan
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Information

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For hydrophilic interaction chromatography

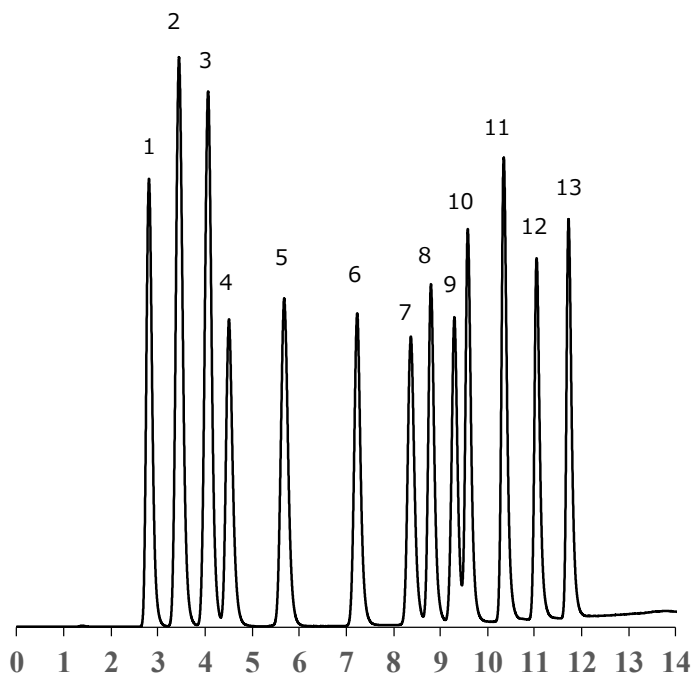
NEW

ULTRON AF-HILIC-CD (HT)

Introducing the AF-HILIC-CD (HT) employing a 2 μm packing material, the latest addition to the AF-HILIC-CD series.

This column can be used for high-throughput analysis of structural homologues and structural isomers.

Simultaneous analysis of mono-, di- and oligosaccharides



- | | |
|--------------|---------------------|
| 1. Xylose | 8. Lactose |
| 2. Fructose | 9. Trehalose |
| 3. Glucose | 10. Xylotriose |
| 4. Mannitol | 11. 1-Kestose (GF2) |
| 5. Xylobiose | 12. Raffinose |
| 6. Sucrose | 13. Nystose (GF3) |
| 7. Lactulose | |

Gradient : 10 minutes gradient, 88% - 65% with 20minutes re-equilibration

Time (min)	Profile	
	%A	%B
0.00	96	4
10.00	58	42
10.01	96	4
30.00	96	4

Column: ULTRON AF-HILIC-CD (HT) (2 μm)
 Column size: 100 mm x 2.0 mm I.D.
 Mobile phase: A 20 mM $\text{CH}_3\text{CO}_2\text{NH}_4$ (pH 6.8) / CH_3CN = 10/90 (v/v)
 B 20 mM $\text{CH}_3\text{CO}_2\text{NH}_4$ (pH 6.8) / CH_3CN = 70/30 (v/v)

Flow rate: 0.26 mL/min
 Column temp.: 40°C
 Detection: ELSD
 Injection vol.: 2.5 μL



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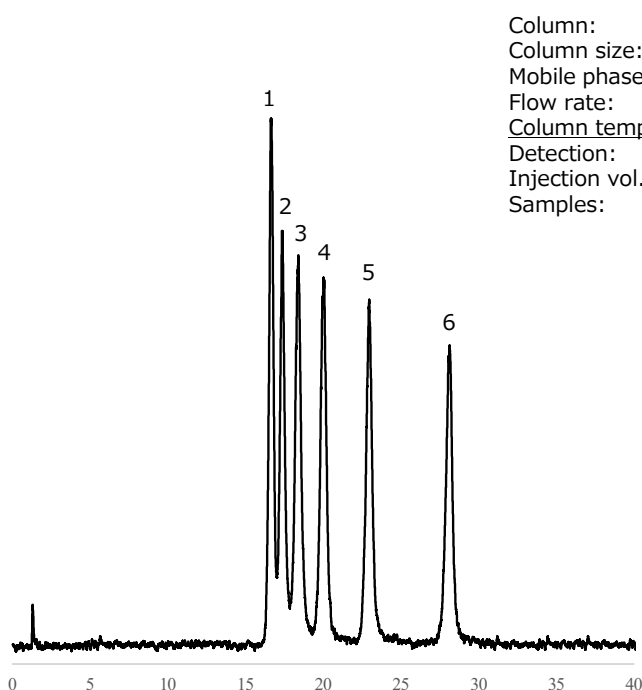
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E-mail: info@shinwa-cpc.co.jp

Website: <http://shinwa-cpc.co.jp/en/>

Separation of sugar isomers



Column: ULTRON AF-HILIC-CD (HT) (2 μm)
 Column size: 150 mm x 2.0 mm I.D.
 Mobile phase: 100 mM $\text{CH}_3\text{CO}_2\text{NH}_4$ (pH 6.8)/ CH_3CN = 10/90 (v/v)
 Flow rate: 0.45 mL/min
 Column temp.: 40°C
 Detection: ELSD
 Injection vol.: 1.0 μL
 Samples:
 1. Turanose (Glucose - Fructose $\alpha(1 \rightarrow 3)$)
 2. Palatinose (Glucose - Fructose $\alpha(1 \rightarrow 6)$)
 3. Maltulose (Glucose - Fructose $\alpha(1 \rightarrow 4)$)
 4. Maltose (Glucose - Glucose $\alpha(1 \rightarrow 4)$)
 5. Lactose (Galactose - Glucose $\beta(1 \rightarrow 4)$)
 6. Trehalose (Glucose - Glucose $\alpha(1 \rightarrow 1)$)

ULTRON AF-HILIC-CD line-up

Product name	Particle size (μm)	Column dimensions Length x I.D. (mm)
ULTRON AF-HILIC-CD (HT)	2	50 x 2.0
		100 x 2.0
		150 x 2.0
ULTRON AF-HILIC-CD	5	100 x 2.0
		100 x 4.6
		150 x 2.0
		150 x 4.6
		250 x 4.6

Please be aware that specifications are subject to change without prior notification.