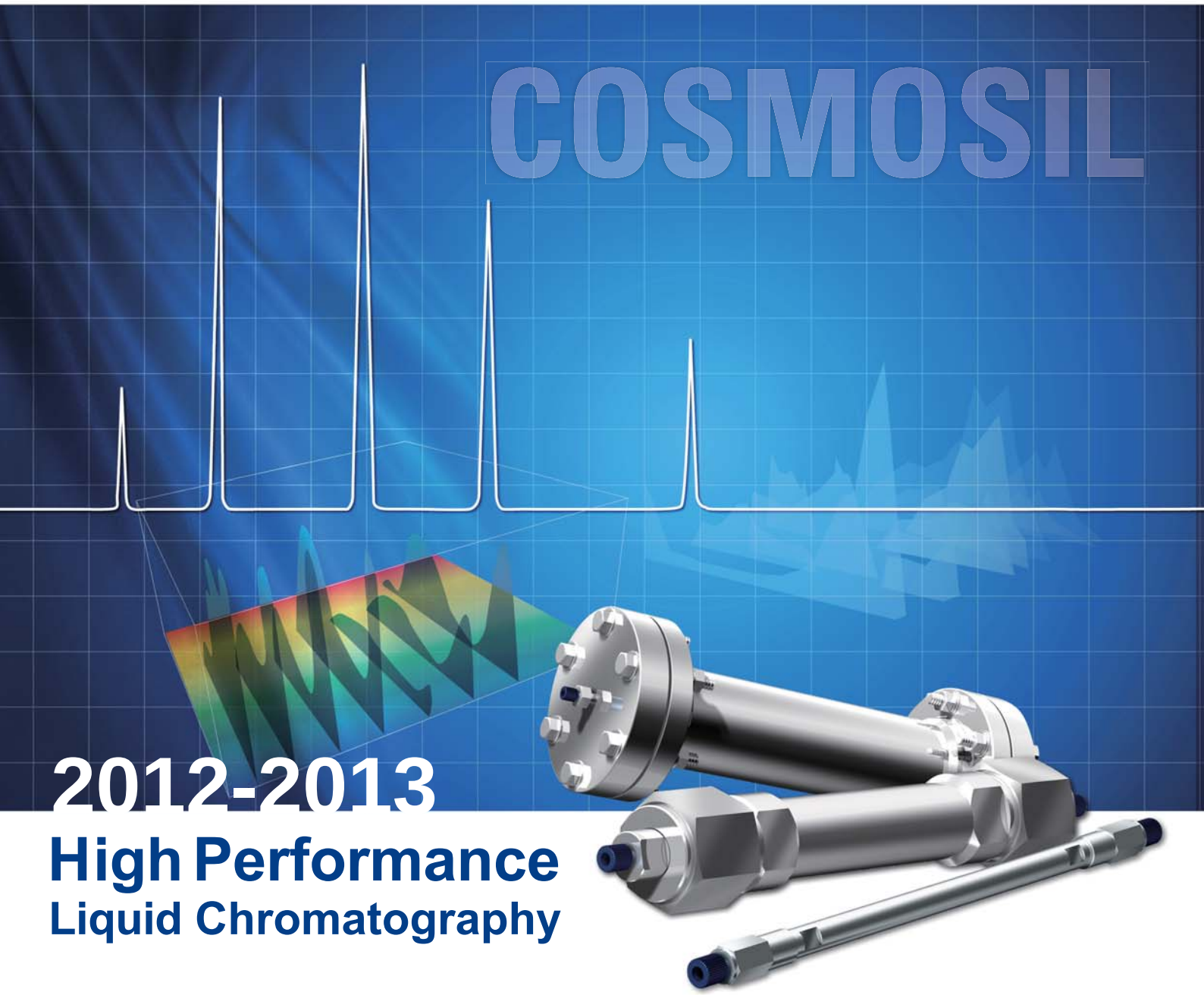
COSMOSIL



2012-2013

High Performance
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COSMOSIL Application has more than 7,000 applications using COSMOSIL columns. Setting optimal HPLC experimental parameters is the one of the most important processes that requires experience and time. COSMOSIL Application provides you with sample analysis conditions with widely used ODS columns and other specialty columns.

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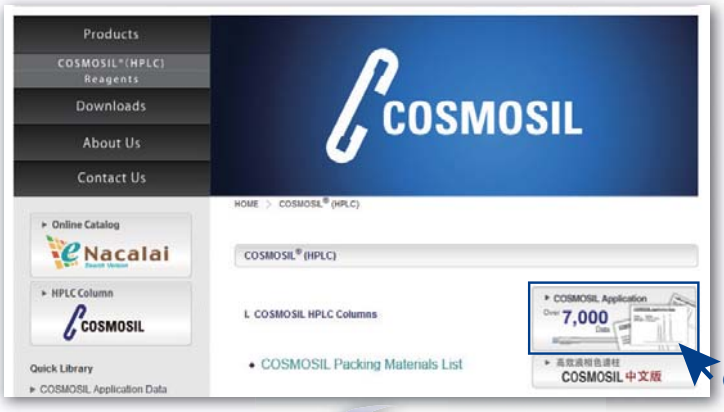

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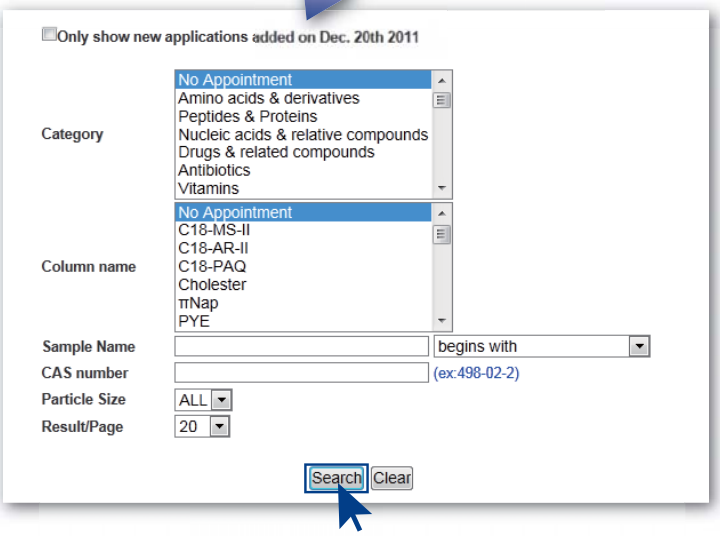
Click



Applications are search by

1. Sample Category
2. Sample Name
3. CAS No.,
4. Column Name
5. Particle Size

Search Result

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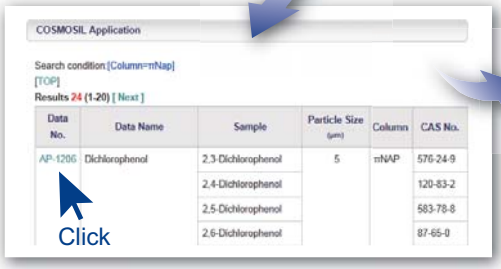
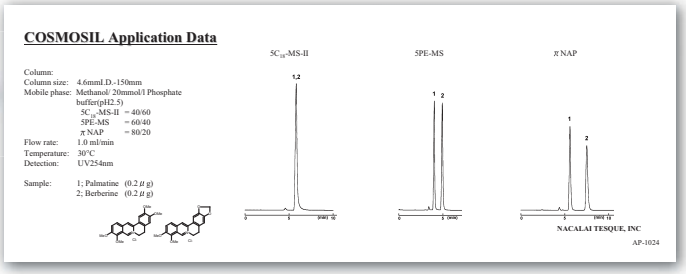


COSMOSIL Application



COSMOSIL/COSMOGEL

Packing Material List

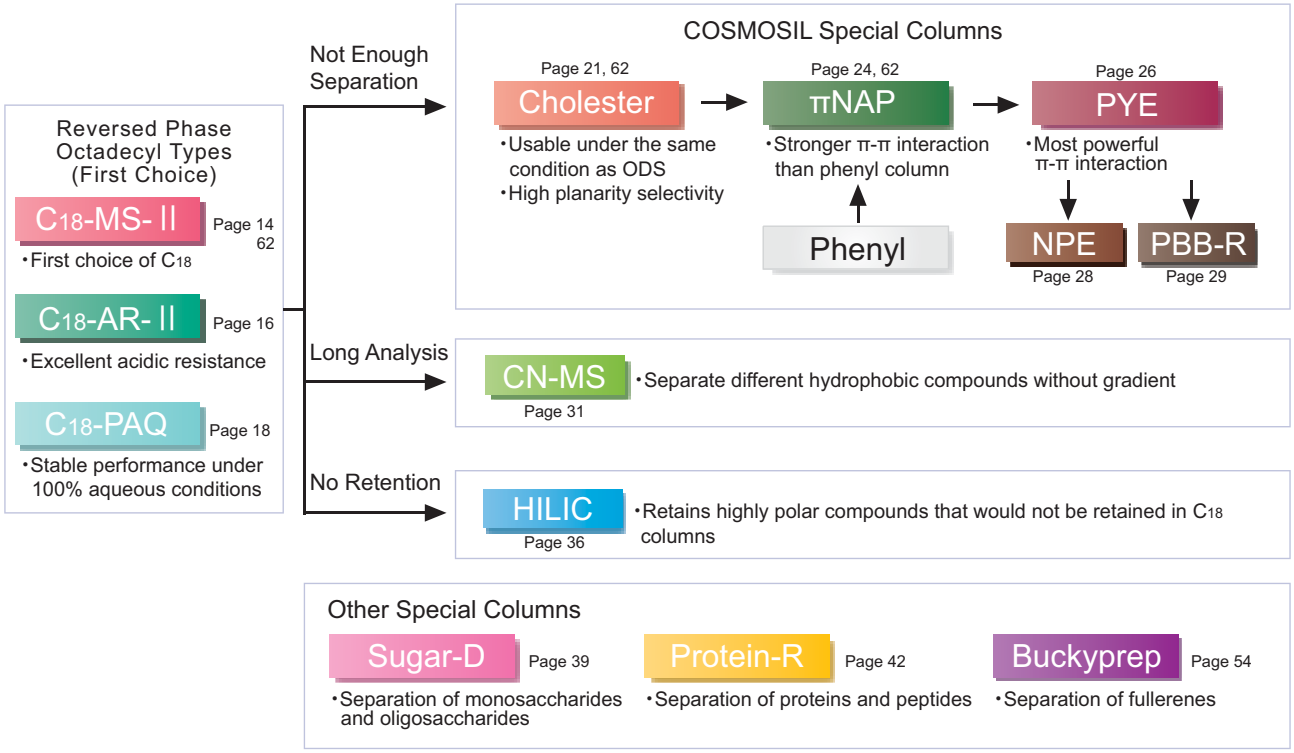
Sample	Separation Mode	Packing Material	Stationary Phase	Special Features and Applications	USP Category	Page
Organic Compunds (low M.W.)	Reversed Phase	C_{18}-MS-II	Octadecyl Group	Multi-purpose C_{18} column. Monofunctional silylation on ultra-pure silica gel for separation of widest range of compounds.	L1	14, 62
		C_{18}-AR-II		Multi-purpose C_{18} column using ultra-pure silica gel. Features strong acid resistance and suitable for a wide range of separation.	L1	16
		C_{18}-PAQ		Reversed phase column, compatible with 100% water based mobile phases.	L1	18
		Cholester	Cholesteryl Group	Usable under the same condition as C_{18}. Unique rigid cholesteryl structure improves separation.		21, 62
		πNAP	Naphthylethyl Group	Stronger π-π interaction than phenyl column		24, 62
		PYE	Pyrenylethyl Group	The most poweful π-π interaction	Coming soon	26
		NPE	Nitrophenylethyl Group	Separation utilizing π-π interaction and Dipole-dipole interaction		28
		PBB-R	Pentabromobenzyl Group	Separation utilizing dispersion force		29
		CN-MS	Cyanopropyl Group	Great reproducibility using isocratic elution mode	L10	31
		C_{22}-AR-II	Docosyl Group	Alkyl chain columns except C_{18} column		32
		C_8-MS	Octyl Group		L7	
		C_4-MS	Butyl Group		L26	
		TMS-MS	Trimethyl Group		L13	
		PE-MS	Phenylethyl Group	π-π interaction	L11	
	Normal Phase	SL-II	--	Normal phase chromatography with non-polar organic solvents	L3	34
	Hydrophilic Interaction	HILIC	Triazole	Retains highly polar compounds that would not be retained in C_{18} column		36
Mono- and Oligosaccharides	Hydrophilic Interaction	Sugar-D	Secondry/Tertiary Amine	A novel stationary phase for saccharide separation. Extended column life and increased stability. Alternative to aminopropyl type		39
		NH_2-MS	Aminopropyl Group	Primary amino bonded column		41
Proteins	Reversed Phase	Protein-R	Octadecyl Group	The most suitable reversed phase column for proteins		42
		C_{18}-AR-300	Octadecyl Group	Wide pore type reversed phase columns with high acid resistance recommended for the separation of proteins, polypeptides, nucleic acids and other large molecules.	L1	44
		C_8-AR-300	Octyl Group		L7	
		C_4-AR-300	Butyl Group		L26	
		Ph-AR-300	Pyrenyl Group		L11	
	Gel Permeation	Diol-120-II	Diol Group	Silica-based gel filtration column for high speed separation of proteins and water soluble polymer	L20	46
		Diol-300-II				
	NEW Ion-exchange	IEX Type Q	Trimethylaminopropyl Type	Anion-exchange Type (purification)		48
		IEX Type Q-N		Anion-exchange Type (ultra-fast analysis, precise analysis)		
		IEX Type S	Sulfopropyl Type	Cation-exchange Type (purification)		
		IEX Type S-N		Cation-exchange Type (ultra-fast analysis, precise analysis)		
		IEX Type M	Trimethylaminopropyl Type/ Sulfopropyl Type	Amphoteric ion-exchange Type (purification)		
		IEX Type M-N		Amphoteric ion-exchange Type (precise analysis)		
	Hydrophobic Interaction	HIC	--	Hydrophobic interaction chromatography column for protein separation		51
Fullerenes	--	Buckyprep	Pyrenylpropyl Group	Standard column for fullerenes separation		54
		Buckyprep-M	Phenothiazinyl Group	Designed to separate metallofullerenes		55
		PBB	Pentabromobenzyl Group	Designed for the preparative separation of fullerenes using carbon disulphide, *o*-dichlorobenzene and toluene		56
		NPE	Nitrophenylethyl Group	Separation of derivatived fullerenes		57
		PYE	Pyrenylethyl Group	Separation of fullerenes and structual isomers		
Carbon Nanotubes	Gel Permeation	CNT-300	Hydrophilic Group (neutral)	Separation of soluble carbon nanotubes		58
		CNT-1000				
		CNT-2000				

For old type columns, please refer to page 59.

COSMOSIL/COSMOGEL Column Chromatography Packing Materials

Separation Mode	Application	Packing Material	Feature	Page
Reversed Phase	Open, Flash, Medium Pressure Chromatography	C_{18}-OPN	Usable under 100% aqueous eluents.	67
		C_{18}-PREP	End-capping treated.	70
Normal Phase		SL-II-PREP	Ultra pure silica gel is used.	71

Column Selection Guide



COSMOSIL USP List

USP No.	Phase	USP Description	Product Name
L01	C_{18}	Octadecyl silane <ODS or C_{18}> chemically bonded to porous silica or ceramic particles, 1.5 to 10 micron in diameter, or a monolithic rod.	COSMOSIL 5C_{18}-MS-II COSMOSIL 5C_{18}-AR-II COSMOSIL 5C_{18}-PAQ COSMOSIL 5C_{18}-AR-300 COSMOSIL 2.5C_{18}-MS-II
L03	SIL	Porous silica particles, 5 to 10 micron in diameter, or a monolithic rod.	COSMOSIL 5SL-II
L07	C_8	Octylsilane <C_8> chemically bonded to porous silica particles, 1.5 to 10 micron in diameter, or a monolithic rod.	COSMOSIL 5C_8-MS COSMOSIL 5C_8-AR-300
L10	CN	Nitrile groups <CN> chemically bonded to porous silica particles, 3 to 10 micron in diameter.	COSMOSIL 5CN-MS
L11	Ph	Phenyl groups chemically bonded to porous silica particles, 1.5 to 10 micron in diameter.	COSMOSIL 5PE-MS COSMOSIL 5Ph-AR-300
L13	C_1	Trimethylsilane <C_1> chemically bonded to porous silica particles, 3 to 10 micron in diameter.	COSMOSIL 5TMS-MS
L20	Diol	Dihydroxypropane groups chemically bonded to porous silica particles, 5 to 10 micron in diameter.	COSMOSIL Diol-120-II COSMOSIL Diol-300-II
L26	C_4	Butyl silane <C_4> chemically bonded to porous silica particles, 3 to 10 micron in diameter.	COSMOSIL 5C_4-MS COSMOSIL 5C_4-AR-300

COSMOSIL Column Size List

Paticle Size 5 µm,15 µm		Length (mm)									
		10	20	30	50	75	100	125	150	250	500
Inner Diameter (mm)	1.0	+	+	+	++	+	++	+	+	+	+
	2.0	+	+	++	++	+	++	+	++	++	+
	3.0	+	+	+	+	+	++	+	++	++	+
	4.6	+	+	++	++	+	++	+	++	++	+
	6.0	+	+	+	+	+	+	+	++	++	+
	8.0	+	+	+	+	+	+	+	+	+	+
	10.0	+	+	+	++	+	+	+	++	++	+
	20.0	+	+	+	++	+	+	+	++	++	+
	28.0	+	+	+	+	+	+	+	+	++	+
	50.0	+	+	+	+	+	+	+	+	++	++

++ Catalog Listed Size
+ Inquire Price and Lead Time

In addition to the original column sizes listed in this catalog, other sizes may be available.
Please contact us at info.intl@nacalai.com for more information.

CORPORATE PROFILE

Nacalai Tesque dates back to 1846 when the company's founder Mansuke Nakarai opened Nakarai Mansuke Shoten, Ltd., an apothecary selling traditional Japanese and Chinese medicines. In 1958, this company's reagent department became an independent company, Nakarai Chemicals, Ltd.

The company has since dedicated itself to expanding its corporate base and has strived to be an enterprise that our customers always rely on, while taking pride in its contribution to scientific and industrial development.

Making the most of this 30-year history and as a step toward the future, Nakarai Chemicals changed its corporate name to Nacalai Tesque, Inc. in 1988. At Nacalai Tesque, we have fostered a corporate commitment to the pursuit of reliable quality and the creation of products of real value, while serving as a vital link between humanity and science. Centering around research chemicals, the fields of our activities include fine chemicals, diagnostics and related laboratory equipment and supplies.

The pace of scientific and technological progress in every industrial field is rapidly accelerating, and all business partners and affiliates are required to provide even more diversified and advanced expertise.

It is our corporate policy to strive for our lofty ideals for excellence while respecting our long history and tradition. We consider it our mission to maintain close contact with our customers by offering reliable quality in all our products, information and services, and by making full use of the knowledge and experience of our staff.

COSMOSIL

INDEX

COSMOSIL

IV. Related Products

V. Applications

VI. Technical Notes

VII. Index

General Information

General Ordering Information

When placing an order with us or making an enquiry, please contact our International Business Development Group or your local distributor. Please clearly identify the product in question when submitting your enquiry. The speed of innovation is accelerating. We always have brand new or improved columns not listed here. There are also many other products Nacalai Tesque can supply. Therefore we urge you to make enquiries.

Product Description and Availability

Please note that the product specifications are subjected to changes and the manufacturing of some product may be stopped. Please consult the table on page 59 for cross-reference information on old products and their newer and better equivalents.

Column Identification

At the end of each section, the COSMOSIL and COSMOGEL packed columns are listed in a way that the particle size, stationary phase, column size of the packing material can be easily determined.

38019-81 COSMOSIL 5 C_{18}-MS-II 4.6 mm I.D. x 150 mm
(1) (2) (3) (4) (5)

When placing an order, please clearly indicate the product number (1), product name (2), particle size (3), type of stationary phase (4) and column size (5).

Warranty Claims

The manufacturer will replace defective columns if notified within 2 weeks of receipt of the product by the customer under the following conditions:

1) Column abnormalities are due to accidents in shipping or rough handling.
2) The number of effective plates of the column is considerably lower than the minimum guaranteed theoretical plate number documented in the inspection report that accompanies each column.

Please contact the International Business Development Section of Nacalai Tesque (info.intl@nacalai.com) or your local distributor for additional information.

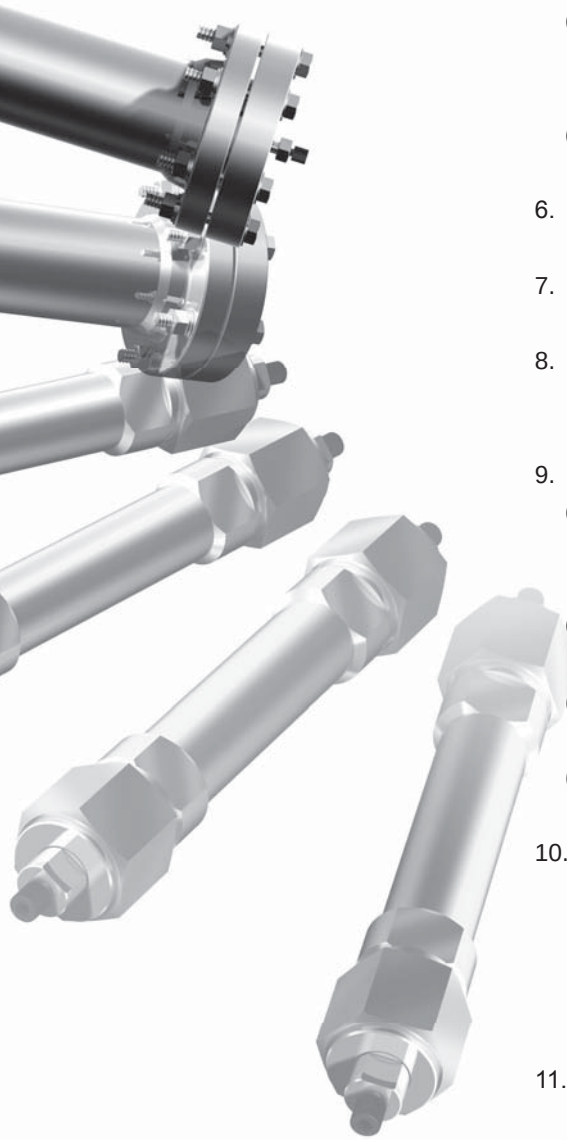
Terms and Conditions of Sale

Terms are subject to conditions set forth by the authorized Nacalai Tesque distributors in each country.

Not for Clinical Use

Nacalai Tesque products are not intended for clinical use. While clinical applications may be shown, these products are not validated for clinical use.

I HPLC Columns

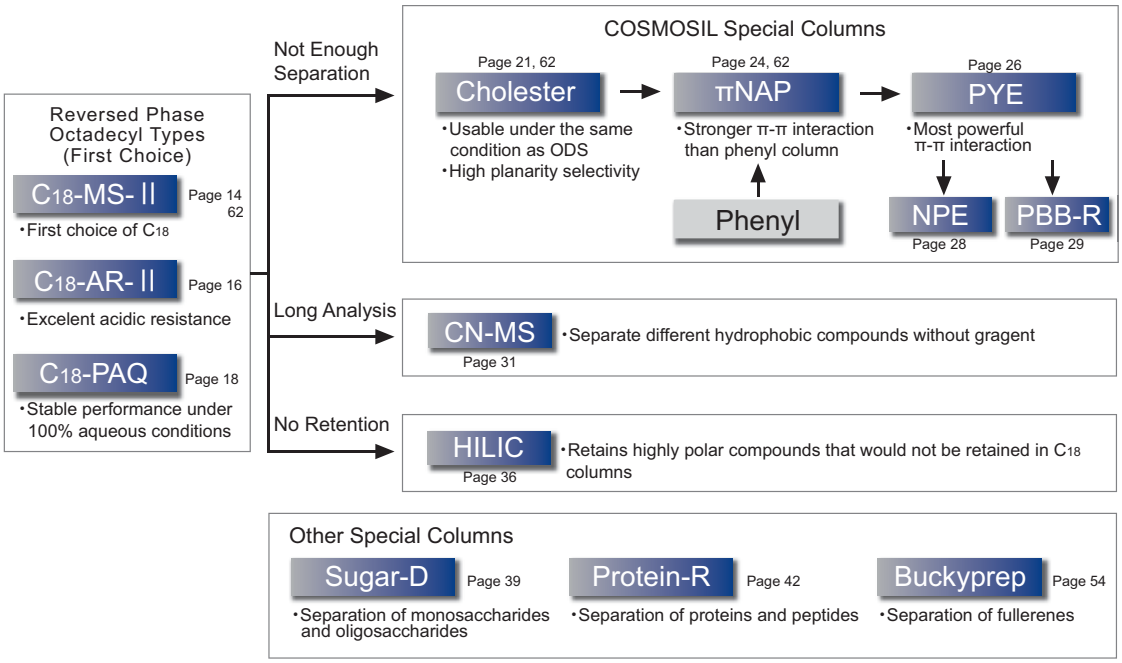


1. COSMOSIL/COSMOGEL HPLC Columns

Sample	Separation Mode	Packing Material	Stationary Phase	Special Features and Applications	USP Category	Page
Organic Compunds (low M.W.)	Reversed Phase	C_{18}-MS-II	Octadecyl Group	Multi-purpose C_{18} column. Monofunctional silylation on ultra-pure silica gel for separation of widest range of compounds.	L1	14, 62
		C_{18}-AR-II		Multi-purpose C_{18} column using ultra-pure silica gel. Features strong acid resistance and suitable for a wide range of separation.	L1	16
		C_{18}-PAQ		Reversed phase column, compatible with 100% water based mobile phases.	L1	18
		Cholester	Cholesteryl Group	Usable under the same condition as C_{18}. Unique rigid cholesteryl structure improves separation.		21, 62
		πNAP	Naphthylethyl Group	Stronger π-π interaction than phenyl column		24, 62
		PYE	Pyrenylethyl Group	The most poweful π-π interaction	Coming soon	26
		NPE	Nitrophenylethyl Group	Separation utilizing π-π interaction and Dipole-dipole interaction		28
		PBB-R	Pentabromobenzyl Group	Separation utilizing dispersion force		29
		CN-MS	Cyanopropyl Group	Great reproducibility using isocratic elution mode	L10	31
		C_{22}-AR-II	Docosyl Group	Alkyl chain columns except C_{18} column		32
		C_8-MS	Octyl Group		L7	
		C_4-MS	Butyl Group		L26	
		TMS-MS	Trimethyl Group		L13	
		PE-MS	Phenylethyl Group	π-π interaction	L11	
	Normal Phase	SL-II	--	Normal phase chromatography with non-polar organic solvents	L3	34
	Hydrophilic Interaction	HILIC	Triazole	Retains highly polar compounds that would not be retained in C_{18} column		36
Mono- and Oligosaccharides	Hydrophilic Interaction	Sugar-D	Secondry/Tertiary Amine	A novel stationary phase for saccharide separation. Extended column life and increased stability. Alternative to amino-propyl type		39
		NH_2-MS	Aminopropyl Group	Primary amino bonded column		41
Proteins	Reversed Phase	Protein-R	Octadecyl Group	The most suitable reversed phase column for proteins		42
		C_{18}-AR-300	Octadecyl Group	Wide pore type reversed phase columns with high acid resistance recommended for the separation of proteins, polypeptides, nucleic acids and other large molecules.	L1	44
		C_8-AR-300	Octyl Group		L7	
		C_4-AR-300	Butyl Group		L26	
		Ph-AR-300	Pyrenyl Group		L11	
	Gel Permeation	Diol-120-II	Diol Group	Silica-based gel filtration column for high speed separation of proteins and water soluble polymer	L20	46
		Diol-300-II				
	NEW Ion-exchange	IEX Type Q	Trimethylaminopropyl Type	Anion-exchange Type (purification)		48
		IEX Type Q-N		Anion-exchange Type (ultra-fast analysis, precise analysis)		
		IEX Type S	Sulfopropyl Type	Cation-exchange Type (purification)		
		IEX Type S-N		Cation-exchange Type (ultra-fast analysis, precise analysis)		
		IEX Type M	Trimethylaminopropyl Type/Sulfopropyl Type	Amphoteric ion-exchange Type (purification)		
		IEX Type M-N		Amphoteric ion-exchange Type (precise analysis)		
	Hydrophobic Interaction	HIC	--	Hydrophobic interaction chromatography column for protein separation		51
Fullerenes	--	Buckyprep	Pyrenylpropyl Group	Standard column for fullerenes separation		54
		Buckyprep-M	Phenothiazinyl Group	Designed to separate metallofullerenes		55
		PBB	Pentabromobenzyl Group	Designed for the preparative separation of fullerenes using carbon disulphide, *o*-dichlorobenzene and toluene		56
		NPE	Nitrophenylethyl Group	Separation of derivatived fullerenes		57
		PYE	Pyrenylethyl Group	Separation of fullerenes and structual isomers		
Carbon Nanotubes	Gel Permeation	CNT-300	Hydrophilic Group (neutral)	Separation of soluble carbon nanotubes		58
		CNT-1000				
		CNT-2000				

For old type columns, please refer to page 59.

2. Column Selection Guide



COSMOSIL Columns Selection Guide

Organic Compounds (low M.W.)

Octadecyl group bonded column (C_{18},ODS) are recommended as first-choice columns for separations of organic compounds (low M.W.). If there is not enough separation or no retention using COSMOSIL C_{18} columns, COSMOSIL series offer many kinds of specialty columns.

- Medicines
- Crude drugs
- Natural compounds
- Pesticides
- Food additives
- Vitamins
- Lipids etc.

Saccharides

- COSMOSIL Sugar-D is recommended for the separation of monosaccharides and oligosaccharides as a first-choice column.
- For the separation of sugar derivatives, COSMOSIL C_{18}-PAQ is suitable as well.

Proteins

- Please select based on the separation mode. Please refer to page 42.

Fullerenes

- COSMOSIL Buckyprep is most suitable for the separation of fullerenes.

3. COSMOSIL Silica Packing Material

Introduction

Superior HPLC columns can be produced only with excellent packing materials and superb packing technique. COSMOSIL columns are well known for their high efficiency and high-resolution separations. Based on spherical, totally porous silica, COSMOSIL columns provide enhanced chemical and mechanical stability as well as very high surface coverage.

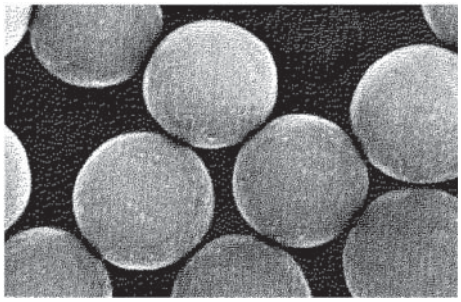
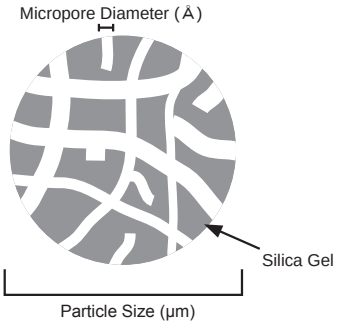
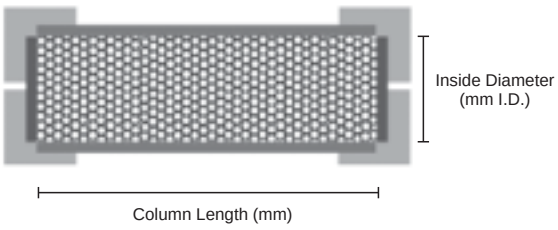


Figure. Microscopic photograph of the silica gel

Packing Material and View Showing a Frame Format of Column





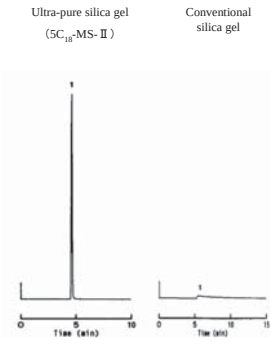
Raw Material Silica Gel

COSMOSIL is based on ultra pure porous spherical silica gel (purity: 99.99% or higher). Low-purity silica gel contains metal impurity which may cause interference in the separation, especially for metal coordination compounds.

Effect of Metal Impurities

Column size: 4.6mmI.D.-150mm
Mobile phase: Acetonitrile/ H_2O = 50/50
Flow rate: 1.0 ml/min
Temperature: 30°C
Detection: UV254nm

Sample: Paeonol



Metal Coordination Compounds

The compounds, which have 2 or more -OH group, -COOH group, -C=O, $-NH_2$ group in the adjacent positions, can complex with metal impurity, which results in peak tailing.

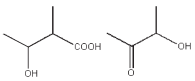


Table. Metal content percentage (ppm)

Packing Material	Metal Content (ppm)						
	Al	Ca	Fe	Mg	Na	Ti	Total
Ultra Pure Silica Gel	4	10	11	3	12	<1	41
Normal Silica Gel	32	450	26	88	56	134	786

Stationary Phase Construction

While C_{18} columns are most widely used in reversed phase HPLC, it is important to distinguish between two very different bonded phase formats. Monomeric type C_{18} format incorporates the bonding of the C_{18} alkyl chain to a single silica atom on the silica gel backbone. Monomeric type columns such as the COSMOSIL C_{18}-MS-II and the MS series have excellent synthesis reproducibility, very good lot-to-lot reproducibility and short mobile phase equilibration times. On the other hand, the polymeric C_{18} format incorporates a tri-functional silylation procedure whereby the octadecyl group is bonded to 2 or 3 silica atoms on the silica gel backbone. This increases silylation results in far greater column stability particularly in acidic mobile phase conditions.

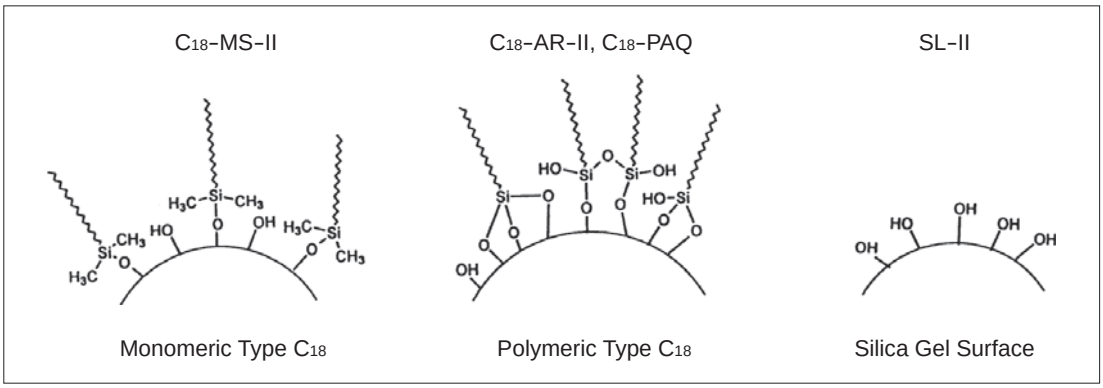
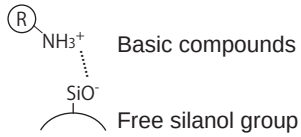


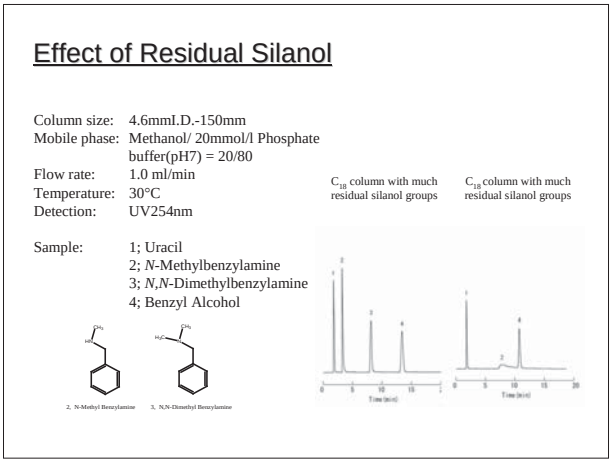
Figure. Diagrams of different stationary phase constructions (before end-capping treatment)

End-capping Treatment

The silanols (Si-OH groups) on the silica surface provided bonding site for stationary phases. However, part of the silanol groups remain un-capped as residual silanol groups even after the end-capping treatment, they cause peak tailing for basic compounds. COSMOSIL packing materials for reversed phase chromatography are of near-perfectly end-capped residual silanol groups.



Basic compound can form ionic bonds with residual free silanols. The ionic bonding causes peak tailing of basic compounds if a silicabased column is not perfectly end-capped.



Synthesis Reproducibility

By using strictly selected silica gel and constant synthesis conditions, the chemically bonded type column retains a variance of the capacity factor (k') between synthetic lots of within ±10% and a variance of the separation factor (α) of within ± 5%. The figures below show in graphic form the lot inspection results of synthesized packing material (COSMOSIL 5C_{18}-MS-II). Figure 1 shows the variance of stationary phase (octadecyl group) introduced volume which is the basic indicator of the quality of the packing material. Figure 2 shows the end-capping efficiency of the packing material. The variance among the lots is reduced to the minimum in the COSMOSIL packed columns.

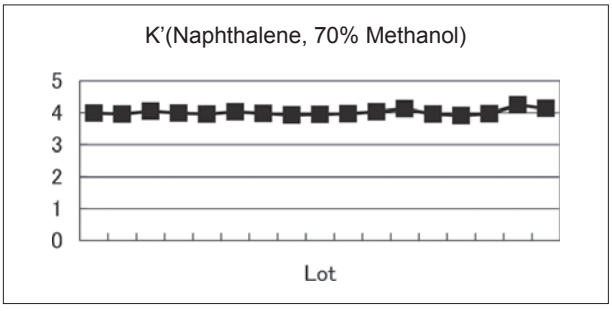


Figure 1. Variance of the combining volume between silica gel and C_{18}

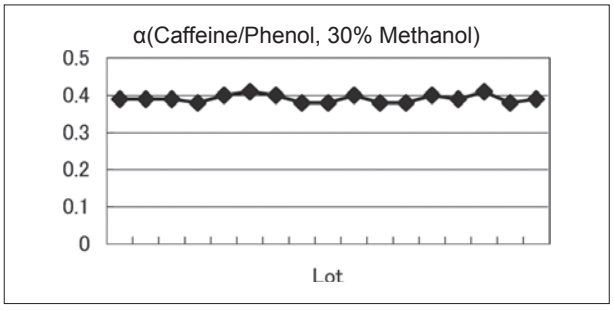


Figure 2. Variance of end capping efficiency of the packing material

4. Performance Guarantee

(1) Quality Guarantee of Packing Materials

Introduction

The strict quality control system of Nacalai Tesque supports the customers with an individual “Inspection Report” which accompanies each and every COSMOSIL and COSMOGEL Packed Column (except guard columns) and an additional “Certificate of Analysis” for the COSMOSIL 5C$_{18}$-MS-II and 5C$_{18}$-AR-II (4.6 mm I.D. x 150 mm and 4.6 mm I.D. x 250 mm).

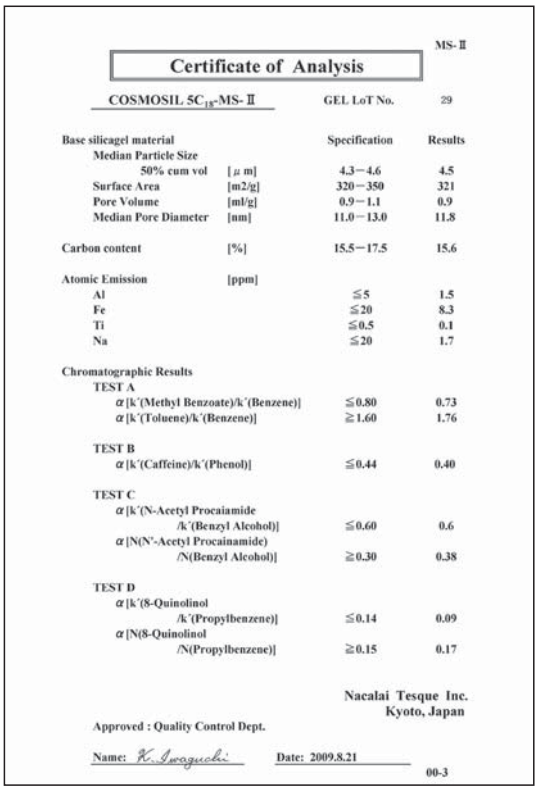
- Validated Columns

Product Name	Product Number	Column Size
COSMOSIL 5C$_{18}$-MS-II	38019-81	4.6 mm I.D. x 150 mm
	38020-41	4.6 mm I.D. x 250 mm
COSMOSIL 5C$_{18}$-AR-II	38144-31	4.6 mm I.D. x 150 mm
	38145-21	4.6 mm I.D. x 250 mm
COSMOSIL Cholester	05976-61	4.6 mm I.D. x 150 mm
	05977-51	4.6 mm I.D. x 250 mm
COSMOSIL HILIC	07056-51	4.6 mm I.D. x 150 mm
	07057-41	4.6 mm I.D. x 250 mm

- COSMOSIL Certificate of Analysis

Validate terms of the physical properties of the silica gel, the carbon content, polar selectivity, hydrophobicity, silanol capacity, steric selectivity, inactive degree to basic and chelating compounds.

E.g.,) Certificate of Analysis (5C$_{18}$-MS-II)



Certificate of Analysis

MS-II

COSMOSIL 5C18-MS-II GEL LoT No. 29

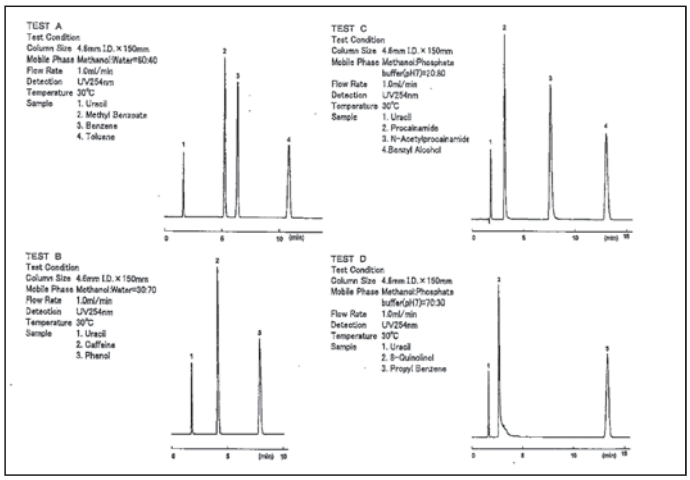
		Specification	Results
Base silicagel material			
Median Particle Size			
50% cum vol	[μm]	4.3–4.6	4.5
Surface Area	[m2/g]	320–350	321
Pore Volume	[ml/g]	0.9–1.1	0.9
Median Pore Diameter	[nm]	11.0–13.0	11.8
Carbon content	[%]	15.5–17.5	15.6
Atomic Emission	[ppm]		
Al		≦5	1.5
Fe		≦20	8.3
Ti		≦0.5	0.1
Na		≦20	1.7
Chromatographic Results			
TEST A			
α[k´(Methyl Benzoate)/k´(Benzene)]		≦0.80	0.73
α[k´(Toluene)/k´(Benzene)]		≧1.60	1.76
TEST B			
α[k´(Caffeine)/k´(Phenol)]		≦0.44	0.40
TEST C			
α[k´(N-Acetyl Procaiamide /k´(Benzyl Alcohol)]		≦0.60	0.6
α[N(N'-Acetyl Procainamide) /N(Benzyl Alcohol)]		≧0.30	0.38
TEST D			
α[k´(8-Quinolinol /k´(Propylbenzene)]		≦0.14	0.09
α[N(8-Quinolinol /N(Propylbenzene)]		≧0.15	0.17

Nacalai Tesque Inc.
Kyoto, Japan

Approved : Quality Control Dept.

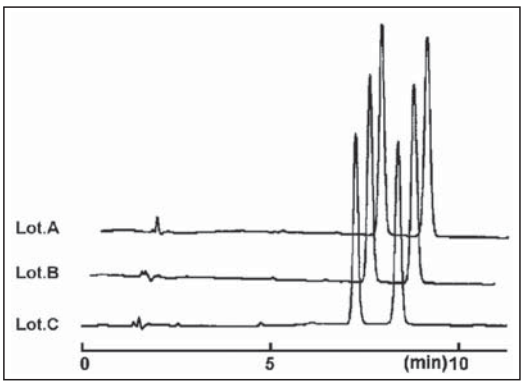
Name: K. Iwaguchi Date: 2009.8.21

00-3



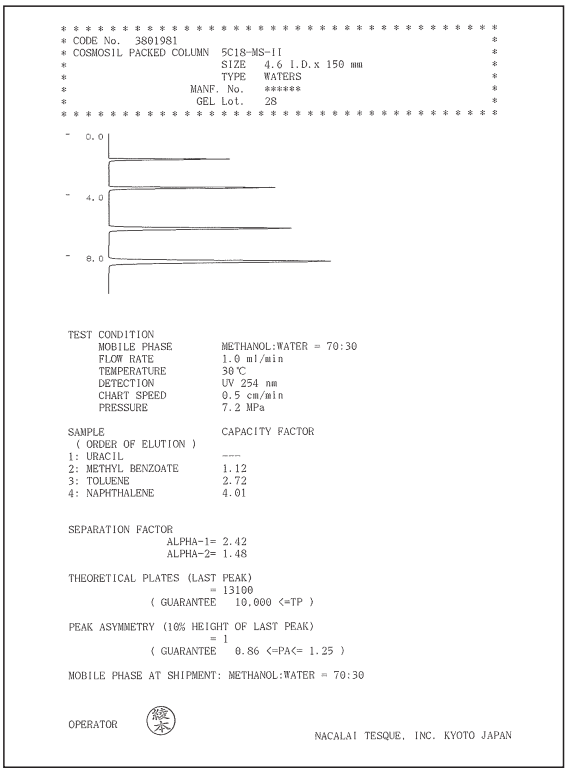
Available in 3 Lots

3 different lots of packing materials are available to demonstrate high reproducibility. Please contact us for more information. (info.intl@nacalai.com)



(2) Quality Guarantee of COSMOSIL Packed Columns

“Inspection Report” is attached to every COSMSOIL Column to guarantee high quality. Please use our strictly controlled products with confidence.



```
* * * * * * * * * * * * * * * * * * * * * * * * * * * * * * * * * * * * *
* CODE No.   3801981                                                     *
* COSMOSIL PACKED COLUMN  5C18-MS-II                                     *
*                         SIZE    4.6 I.D.x 150 mm                      *
*                         TYPE    WATERS                                *
*                    MANF. No.    ******                                *
*                     GEL Lot.    28                                    *
* * * * * * * * * * * * * * * * * * * * * * * * * * * * * * * * * * * * *
```



```
TEST CONDITION
     MOBILE PHASE          METHANOL:WATER = 70:30
     FLOW RATE             1.0 ml/min
     TEMPERATURE           30℃
     DETECTION             UV 254 nm
     CHART SPEED           0.5 cm/min
     PRESSURE              7.2 MPa

SAMPLE                     CAPACITY FACTOR
  ( ORDER OF ELUTION )
1: URACIL                  ---
2: METHYL BENZOATE         1.12
3: TOLUENE                 2.72
4: NAPHTHALENE             4.01

SEPARATION FACTOR
                ALPHA-1= 2.42
                ALPHA-2= 1.48

THEORETICAL PLATES (LAST PEAK)
                   = 13100
             ( GUARANTEE   10,000 <=TP )

PEAK ASYMMETRY (10% HEIGHT OF LAST PEAK)
                   = 1
             ( GUARANTEE   0.86 <=PA<= 1.25 )

MOBILE PHASE AT SHIPMENT: METHANOL:WATER = 70:30

OPERATOR                                  NACALAI TESQUE, INC. KYOTO JAPAN
```

Sample of Inspection Report

Inspection Items

- Theoretical plate number
- Peak asymmetry
- Capacity factor
- Separation factor
- Pressure

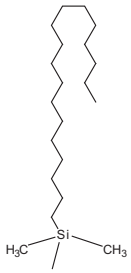
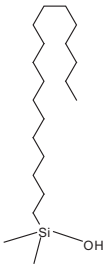
5. Reversed Phase Chromatography Columns

(1) C_{18} (ODS) Series

Introduction

The reversed phase HPLC column is most commonly used because of the high theoretical plate number, excellent separation characteristics, reproducibility, affordable cost and ease of use. Columns packed with the octadecyl group bonded type silica gel (C_{18}, ODS) are the most widely employed. We offer three types of octadecyl group bonded columns: COSMOSIL C_{18}-MS-II, C_{18}-AR-II and C_{18}-PAQ, each of which has a different separation property.

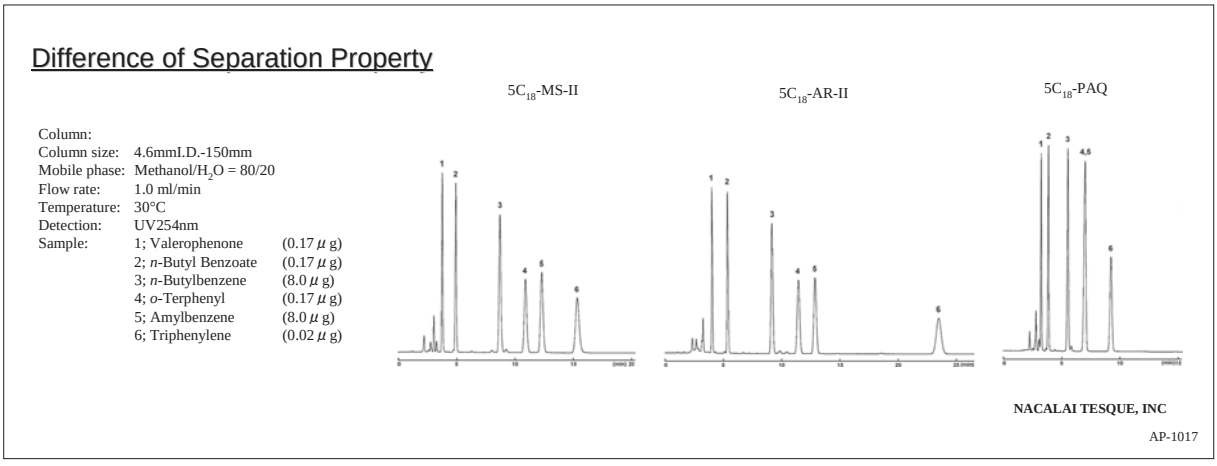
Specifications

Packing Material	C_{18}-MS-II	C_{18}-AR-II	C_{18}-PAQ
Silica Gel	High Purity Porous Spherical Silica		
Average Particle Size	3 , 15μm*	3, 5, 15 μm	5, 15 μm
Average Pore Size	approx. 120 Å		
Specific Surface Area	approx. 300 m^2/g		
Bonded Phase Stracture			
Bonded Phase	Octadecyl Group		
Bonding Type	Monomeric	Polymeric	
Main Interaction	Hydrophobic Interaction		
End-capping Treatment	Near-perfect Treatment		
Carbon Load	approx. 16%	approx. 11%	approx. 11%
Usable pH Range	2~10**	1.5~7.5**	2~7.5
Features	・Multi-purpose C_{18} Column ・Suitable for basic compounds	・Features strong acid resistance ・Suitable for acid compounds and peptides	・Suitable for hydrophilic compounds. ・Compatible with 100% water based mobile phase.

* For 2.5C18-MS-II (2.5 μm), please refer to page 62.
**Optimal pH range of silica-based columns is between 2 and 7.5. Extreme pH may significantly decrease column lifetime.

Difference of Separation Property

Comparing to COSMOSIL 5C_{18}-MS-II;
COSMOSIL 5C_{18}-AR-II retains planar structure compouds (Sample 6) longer. COSMOSIL 5C_{18}-PAQ has shorter retention time, and retains polar compounds (Sample 1,2) longer.

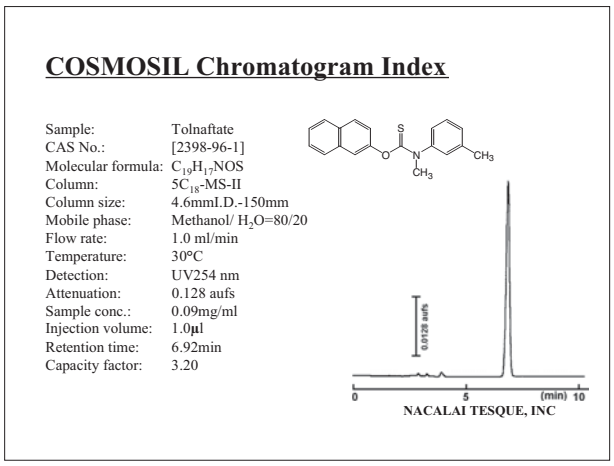


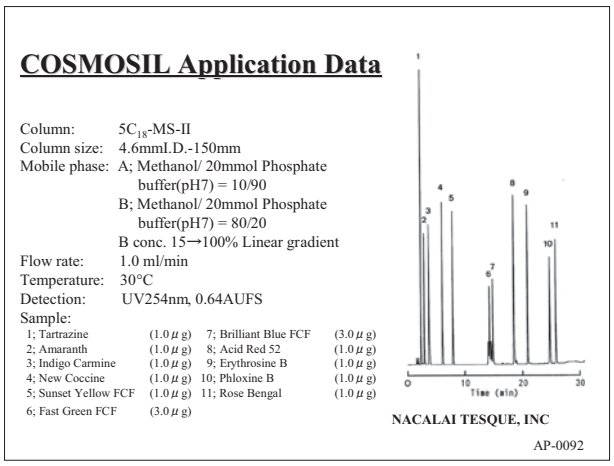
Column Selection Based on Applications

We prepare the following application data to help you select separation conditions.

COSMOSIL Application

COSMOSIL Application has more than 7,000 applications using COSMOSIL columns. Setting optimal HPLC experimental parameters is the one of the most important processes that requires experience and time. COSMOSIL Application provides you with sample analysis conditions with widely used ODS columns and other specialty columns. For more information, please visit COSMSOIL Application page on our website at http://www.nacalai.co.jp/global/cosmosil/. For more information, please see page 90.





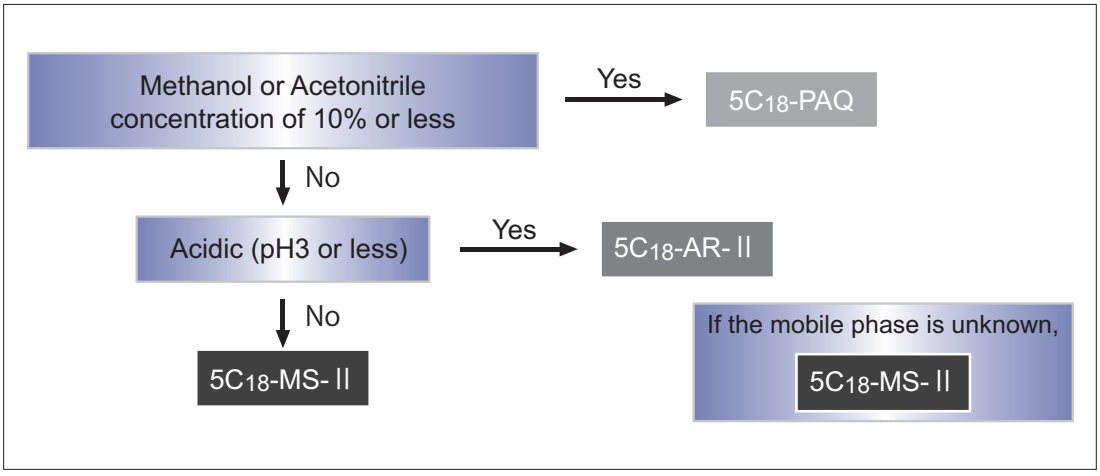
Applications of Drug Substances in the Japanese Pharmacopeia, 15th

We have prepared drug analysis HPLC data using three different COSMOSIL C18 columns as specified in Japanese Pharmacopoeia, 15th version. 5um silica gel column size 4.6mm I.D. x 150 mm or 250 mm were used with reagent grade samples. Furthermore, we analyzed reference drug substances with internal standard and impurities. The data are available at our web site at http://www.nacalai.co.jp/global/cosmosil/, or search “COSMOSIL Japanese Pharmacopoeia” on the web. An example of the application is shown after page 91.

Column Selection by Mobile Phase

- If a mobile phase is determined, use the following chart to select an appropriate COSMOSIL column.
- Refer to application above for choosing a mobile phase of new analysis.
- Adjustment of pH is required for dissociative compounds.
- Generally acidic mobile phase is suitable for acidic compounds, and neutral mobile phase is suitable for basic compounds.
- If you are not sure about the mobile phase, try C18-MS-II first.

Column Selection Guide by Mobile Phase



COSMOSIL C_{18}-MS-II

- Multi-purpose C_{18} Column
- High reproducibility
- A wide range of applications

Separation Property

The COSMOSIL 5C_{18}-MS-II is a well-balanced column with better basic performance such as sharper peaks of basic compounds and chelating compounds, large hydrophobic interaction, low analytical pressure, and high theoretical plate number. The COSMOSIL 5C_{18}-MS-II is first-choice column for reversed phase chromatography.

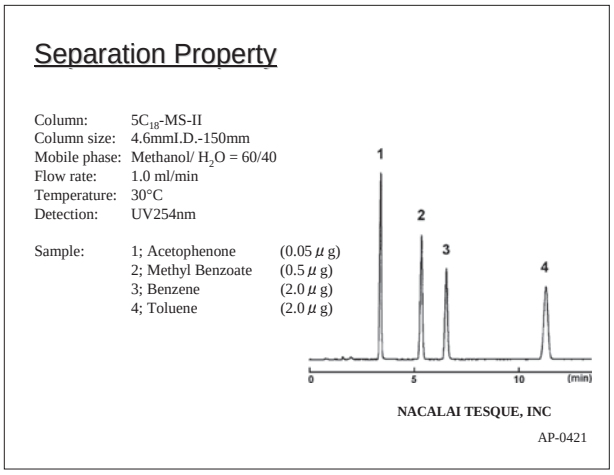
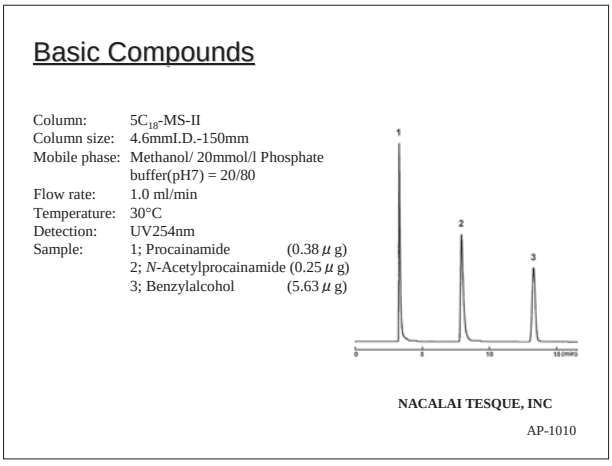


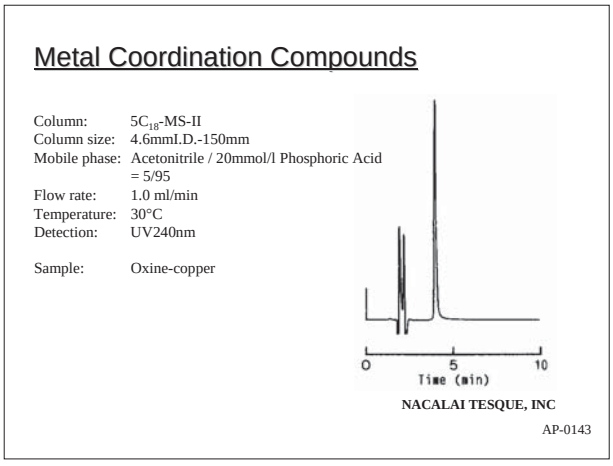
Table. Comparison of hydrophobic interaction, analytical pressure, and theoretical plate number

Column	Hydrophobic Interaction α (Toluene/Benzene)	Pressure (MPa)	Theoretical Plate Number (Toluene)
COSMOSIL 5C_{18}-MS-II	1.96	8.3	14300
A Company C_{18}	1.99	13.0	16800
B Company C_{18}	1.94	8.0	14000
C Company C_{18}	1.69	11.2	5600
D Company C_{18}	1.84	10.5	14200

Analysis of Basic Compounds and Metal Coordination Compounds

The COSMOSIL 5C_{18}-MS-II column, taking advantage of a new end-capping treatment, can replace the original COSMOSIL C_{18} (ODS) column. A new end-capping treatment with polar groups for "shield effect" has significantly improved peak shape for basic compounds. Ultra pure silica gel with low trace-metal content is used for COSMOSIL columns; thus the columns provide excellent peak shapes for chelating compounds.





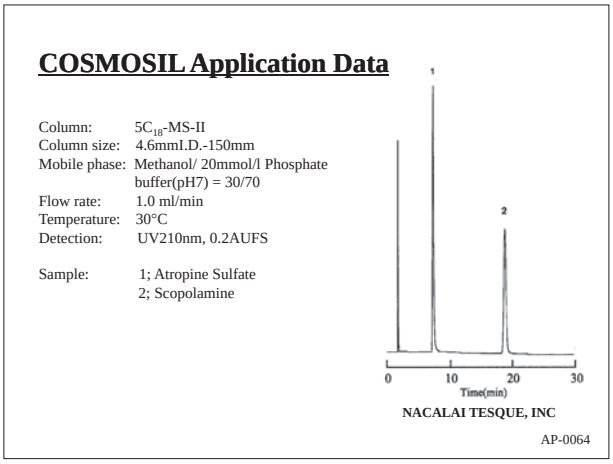
High Reproducibility

The strict quality control system of Nacalai Tesque supports the customers with an individual “Inspection Report” which accompanies each and every COSMOSIL and COSMOGEL Packed Column (except guard columns) and an additional “Certificate of Analysis” for the COSMOSIL 5C$_{18}$-MS-II (4.6 mm I.D. x 150 mm and 4.6 mm I.D. x 250 mm). For more informations, please refer to page 10.

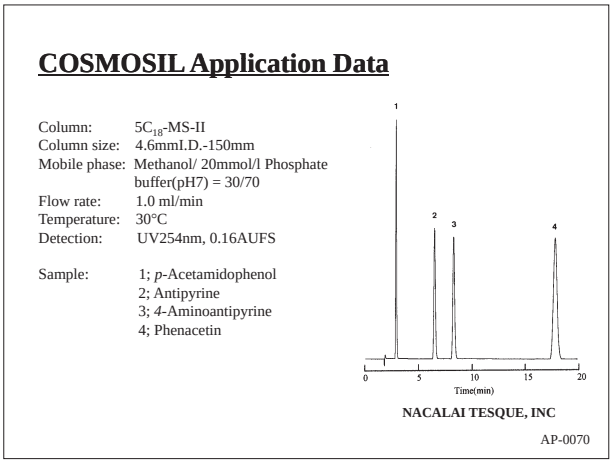
A Wide Range of Applications

A wide selection of applications, e.g. drug molecules, is available to achieve appropriate separation parameters for target samples.

● Parasympatholytic Agents



● Analgesic Antipyretic Drugs



Ordering Information

● Analytical / Preparative Column (Particle Size: 5 µm)

COSMOSIL 5C$_{18}$-MS-II Packed Column

Column Size I.D. x Length (mm)	Product Number
1.0 x 50	02824-31
1.0 x 150	02896-01
2.0 x 30	05876-71
2.0 x 50	04355-21
2.0 x 100	05597-31
2.0 x 150	38025-91
2.0 x 250	05761-61
3.0 x 100	05458-51
3.0 x 150	34245-31
3.0 x 250	34254-11
4.6 x 30	34341-61
4.6 x 50	38017-01

Column Size I.D. x Length (mm)	Product Number
4.6 x 100	38018-91
4.6 x 150	38019-81
4.6 x 150 3 lots set*	09397-73
4.6 x 250	38020-41
6.0 x 150	38021-31
6.0 x 250	38022-21
10 x 50	05789-21
10 x 150	34355-91
10 x 250	38023-11
20 x 150	05091-41
20 x 250	38024-01
28 x 250	05760-71

COSMOSIL 5C$_{18}$-MS-II Guard Column

Column Size I.D. x Length (mm)	Product Number
4.6 x 10	38014-31
4.6 x 10 Cartridge**	38015-89
10 x 20	38016-11
20 x 20	05790-81
20 x 50	34371-71
28 x 50	34347-01

* For 4.6 x 150 3 lots set, please refer to page 11.
** 3 cartridges included, needs a holder, refer to page 87.

● Preparative Column (Particle Size: 15 µm)

COSMOSIL 15C$_{18}$-MS-II Packed Column

Column Size I.D. x Length (mm)	Product Number
28 x 250	34525-61
50 x 250	05886-41
50 x 500	34531-71

COSMOSIL 15C$_{18}$-MS-II Guard Column

Column Size I.D. x Length (mm)	Product Number
28 x 50	05885-51
50 x 50	34527-41

● Fast LC Column (Particle Size: 3 µm)

COSMOSIL 3C$_{18}$-MS-II Packed Column

Column Size I.D. x Length (mm)	Product Number
2.0 x 50	05514-01
4.6 x 10	38065-71
4.6 x 50	38066-61
4.6 x 100	38067-51

For more information, please refer to page 33 for 15C$_{18}$-MS-II (15 µm) and page 62 for 2.5C$_{18}$-MS-II (2.5 µm).
For flow rate and equipment of semi-micro columns, or scale up to preparative columns, please refer to page 189.
For 5C$_{18}$, 5C$_{18}$-MS, please refer to page 59.

COSMOSIL C_{18}-AR-II

- Features strong acid resistance
- Suitable for acid compounds and peptides

Acid Resistance

The COSMOSIL 5C_{18}-AR-II packed column features a polymeric type of C_{18} reversed phase material. The acidic resistance of COSMOSIL 5C_{18}-AR-II is much improved compared with commercially available monomeric type octadecyl stationary phases. It retains high performance even in case of acidic mobile phases commonly used to separate acidic compounds and peptides.

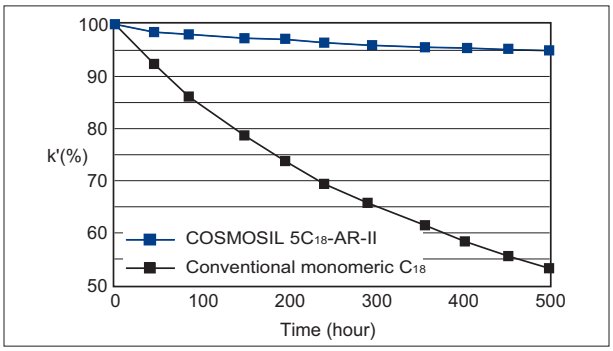


Figure.
Decomposition test in 0.1% Trifluoroacetic acid solution at 60°C.
Capacity factor (k') = Naphthalene,
Mobile phase: Methanol / H_2O=70/30

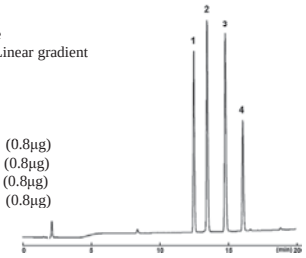
Applications

- Peptides

COSMOSIL Application Data

Column: 5C_{18}-AR-II
Column size: 4.6mmI.D.-150mm
Mobile phase: A; 0.05%TFA-H_2O
B; 0.05%TFA-Acetonitrile
B conc. 10→40% 20min Linear gradient
Flow rate: 1.0 ml/min
Temperature: 30°C
Detection: UV220nm

Sample:
1; Oxytocin (0.8µg)
2; Angiotensin II(Human) (0.8µg)
3; Angiotensin I(Human) (0.8µg)
4; Substance P (0.8µg)



NACALAI TESQUE, INC

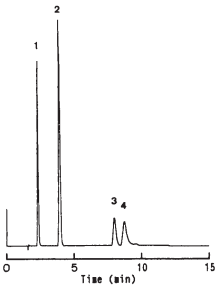
AP-0351

- Organic Acids

COSMOSIL Application Data

Column: 5C_{18}-AR-II
Column size: 4.6mmI.D.-150mm
Mobile phase: Methanol/ 20mmol/l Phosphoric Acid = 20/80
Flow rate: 1.0 ml/min
Temperature: 30°C
Detection: UV254nm, 0.5AUFS

Sample:
1; Gallic Acid (0.63µg)
2; Protocatechuic Acid (0.63µg)
3; Gentisic Acid (0.63µg)
4; Phthalic Acid (0.63µg)



NACALAI TESQUE, INC

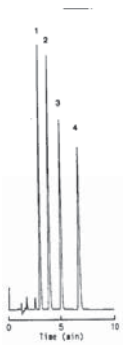
AP-0159

- Salicylic Acid Esters

COSMOSIL Application Data

Column: 5C_{18}-AR-II
Column size: 4.6mmI.D.-150mm
Mobile phase: Acetonitrile/ H_2O = 70/30
Flow rate: 1.0 ml/min
Temperature: 30°C
Detection: UV254nm, 0.2AUFS

Sample:
1; Methyl Salicylate (2.3µg)
2; Ethyl Salicylate (2.6µg)
3; *n*-Propyl Salicylate (2.3µg)
4; *n*-Butyl Salicylate (2.6µg)



NACALAI TESQUE, INC

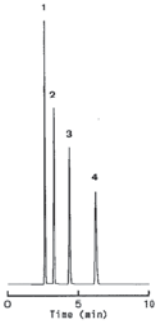
AP-0165

- Parabens

COSMOSIL Application Data

Column: 5C_{18}-AR-II
Column size: 4.6mmI.D.-150mm
Mobile phase: Acetonitrile/ H_2O = 50/50
Flow rate: 1.0 ml/min
Temperature: 30°C
Detection: UV254nm, 0.12AUFS

Sample:
1; Methyl *p*-Hydroxybenzoate (0.125µg)
2; Ethyl *p*-Hydroxybenzoate (0.125µg)
3; *n*-Propyl *p*-Hydroxybenzoate (0.125µg)
4; Butyl *p*-Hydroxybenzoate (0.125µg)

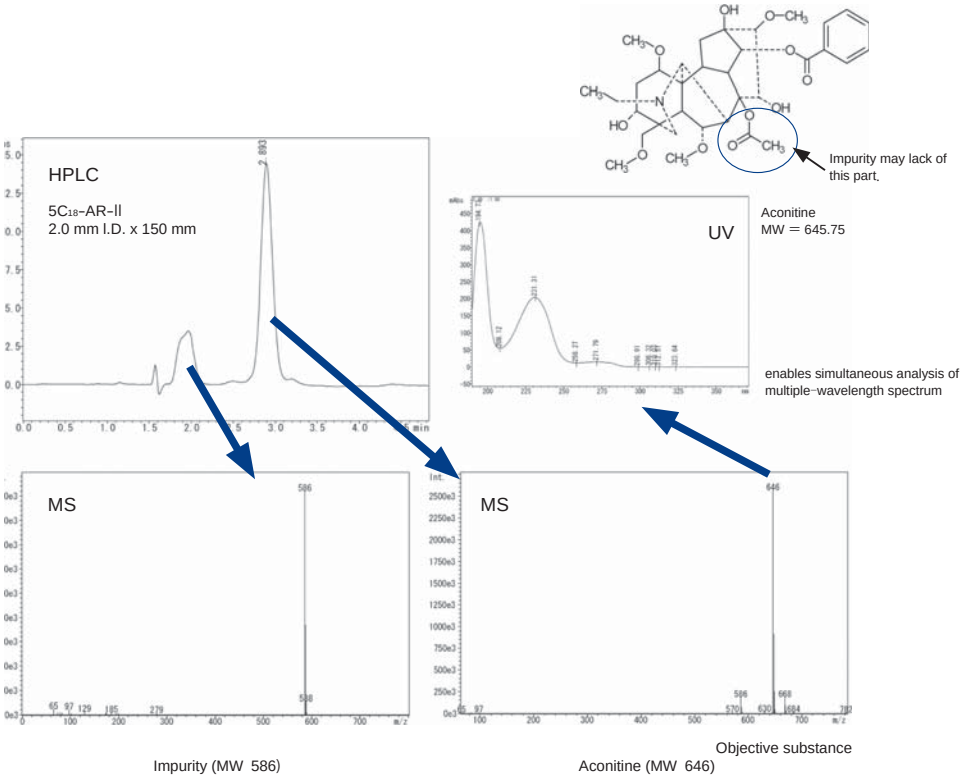


NACALAI TESQUE, INC

AP-0099

LC/MS Applications

● Identification of herbal medicine constituents by LC/MS



Ordering Information

● Analytical / Preparative Column (Particle Size: 5 µm)

COSMOSIL 5C18-AR-II Packed Column

Column Size I.D. x Length (mm)	Product Number
1.0 x 50	02955-21
1.0 x 150	02951-61
2.0 x 30	05098-71
2.0 x 50	34400-81
2.0 x 100	34469-11
2.0 x 150	37992-51
2.0 x 250	05272-71
3.0 x 100	05791-71
3.0 x 150	38028-61
3.0 x 250	38029-51
4.6 x 30	05877-61
4.6 x 50	38142-51
4.6 x 100	38143-41
4.6 x 150	38144-31
4.6 x 150 3 lots set*	09396-83
4.6 x 250	38145-21
6.0 x 150	38146-11
6.0 x 250	38147-01
10 x 50	05369-21
10 x 150	34350-41
10 x 250	38149-81
20 x 150	34316-01
20 x 250	38150-41
28 x 250	34362-91

COSMOSIL 5C18-AR-II Guard Column

Column Size I.D. x Length (mm)	Product Number
4.6 x 10	38141-61
4.6 x 10 Cartridge**	38008-89
10 x 20	38148-91
20 x 20	34458-51
20 x 50	34479-81
28 x 50	34363-81

*For 4.6 x 150 3 lots set, please refer to page 11.
**3 cartridges included, needs a holder, refer to page 87.

● Preparative Column (Particle Size: 15 µm)

COSMOSIL 15C18-AR-II Packed Column

Column Size I.D. x Length (mm)	Product Number
28 x 250	37978-51
50 x 250	38058-71
50 x 500	05884-61

COSMOSIL 15C18-AR-II Guard Column

Column Size I.D. x Length (mm)	Product Number
28 x 50	38030-11
50 x 50	38057-81

COSMOSIL 3C18-AR-II Packed Column

Column Size I.D. x Length (mm)	Product Number
2.0 x 50	05478-91
4.6 x 10	38068-41
4.6 x 50	38069-31
4.6 x 100	38070-91

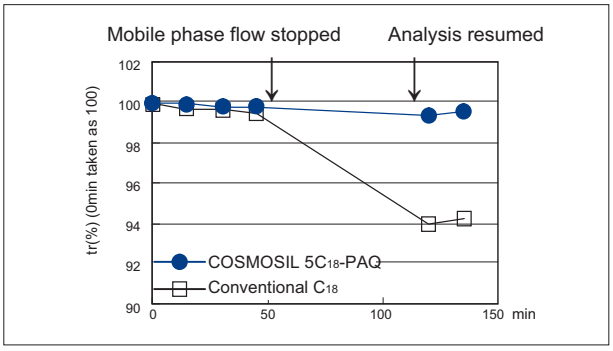
For 15C18-AR-II, please refer to page 33.
For flow rate and equipment of semi-micro columns, or scale up to preparative columns, please refer to page 189.
For 5C18, 5C18-AR, please refer to page 59.

COSMOSIL C_{18}-PAQ

- Compatible with 100% water based mobile phase
- Suitable for hydrophilic compounds

Stable Performance

Stable performance under 100% aqueous conditions



The figure shows the change of retention time for thymine with 100% aqueous mobile phase (20 mmol/l phosphate buffer pH7). The sample was analyzed 4 times (1 hour). Flow of mobile phase was then stopped for 1 hour. The sample was analyzed under the same condition again after 1 hour. The conventional C_{18} column showed change of retention time, but COSMOSIL 5C_{18}-PAQ maintained stable retention time.

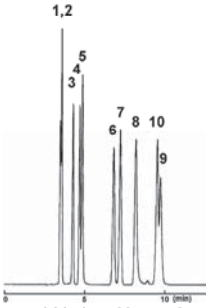
Applications

- Organic Acids

COSMOSIL Application Data

Column: 5C_{18}-PAQ
Column size: 4.6mmI.D.-250mm
Mobile phase: 20mmol/l Phosphate buffer(pH2.5)
Flow rate: 1.0 ml/min
Temperature: 30°C
Detection: UV210nm

Sample:
1; Glycolic Acid (6.3µg)
2; Tartaric Acid (4.0µg)
3; *L*-(-)-Malic Acid (6.4µg)
4; Lactic Acid (13µg)
5; Acetic Acid (13µg)
6; Citric Acid(6.3µg)
7; Succinic Acid (13µg)
8; Maleic Acid (0.06µg)
9; Propionic Acid (13µg)
10; Fumaric Acid (0.03µg)



NACALAI TESQUE, INC

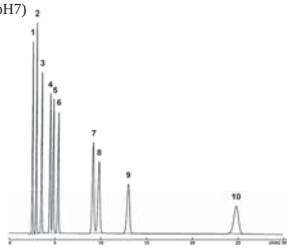
AP-1058

- Nucleobases and Nucleosides

COSMOSIL Application Data

Column: 5C_{18}-PAQ
Column size: 4.6mmI.D.-150mm
Mobile phase: 20mmol/l Phosphate buffer(pH7)
Flow rate: 1.0 ml/min
Temperature: 30°C
Detection: UV260nm

Sample:
1; Cytosine (0.16µg)
2; Uracil (0.16µg)
3; Cytidine (0.32µg)
4; Uridine (0.24µg)
5; Guanine (0.16µg)
6; Thymine (0.16µg)
7; Adenine (0.08µg)
8; Guanosine (0.24µg)
9; Thymidine (0.24µg)
10; Adenosine (0.16µg)



NACALAI TESQUE, INC

AP-0375

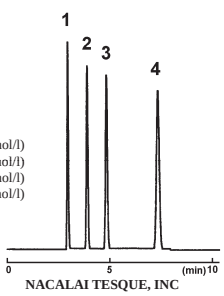
- dNTP

COSMOSIL Application Data

Column: 5C_{18}-PAQ
Column size: 4.6mmI.D.-250mm
Mobile phase: 20mmol/l Phosphate buffer(pH7)
Flow rate: 1.0 ml/min
Temperature: 30°C
Detection: UV260nm, 0.16AUFS

Sample:
1; 2'-Deoxycytidine 5'-Triphosphate [dCTP] (1mmol/l)
2; 2'-Deoxythymidine 5'-Triphosphate [dTTP] (1mmol/l)
3; 2'-Deoxyguanosine 5'-Triphosphate [dGTP] (1mmol/l)
4; 2'-Deoxyadenosine 5'-Triphosphate [dATP] (1mmol/l)

Injection 0.5µl



NACALAI TESQUE, INC

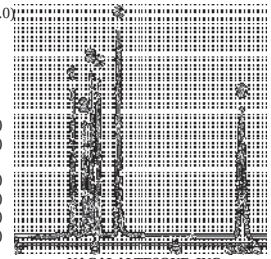
AP-0371

- Catecholamines

COSMOSIL Application Data

Column: 5C_{18}-PAQ
Column size: 4.6mmI.D.-250mm
Mobile phase: 20mmol/l Phosphate buffer(pH7.0)
Flow rate: 1.0 ml/min
Temperature: 30°C
Detection: UV270nm

Sample:
1; *L*-Noradrenaline (0.38µg)
2; (±)-Epinephrine (0.38µg)
3; *DL*-3-Methoxy-4-hydroxymandelic Acid (0.56µg)
4; *L*-DOPA (0.56µg)
5; Dopamine (1.13µg)
6; Homovanillic Acid (1.13µg)



NACALAI TESQUE, INC

AP-1056

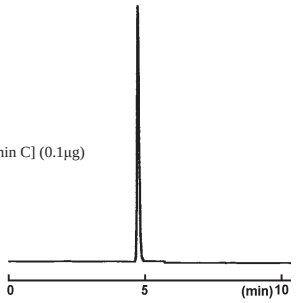
Applications

● Ascorbic Acid

COSMOSIL Application Data

Column: $5C_{18}$-PAQ
Column size: 4.6mmI.D.-250mm
Mobile phase: 20mmol/l Phosphoric Acid
Flow rate: 1.0 ml/min
Temperature: 30°C
Detection: UV245nm, 0.16AUFS

Sample: *L*(+)-Ascorbic Acid [Vitamin C] (0.1µg)



NACALAI TESQUE, INC

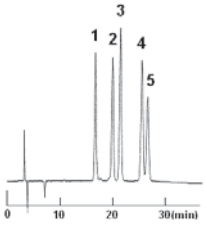
AP-0372

● *2*-Phenylethyl glycosides

COSMOSIL Application Data

Column: $5C_{18}$-PAQ
Column size: 4.6mmI.D.-150mm
Mobile phase: Acetonitrile/ Methanol/ H_2O = 8/4/8
Flow rate: 1.0 ml/min
Temperature: 30°C
Detection: UV210nm

Sample: 1; *2*-Phenylethyl-β-melibioside
2; *2*-Phenylethyl-β-gentiobioside
3; *2*-Phenylethyl-β-lactoside
4; *2*-Phenylethyl-β-cellobioside
5; *2*-Phenylethyl-β-maltoside



NACALAI TESQUE, INC

Data courtesy of Dr. K. Sakata, Dr. B. Shimizu, Institute for Chemical Research, Kyoto University

Ordering Information

● Analytical / Preparative Column (Particle Size: 5 µm)

COSMOSIL $5C_{18}$-PAQ Packed Column

Column Size I.D. x Length (mm)	Product Number
1.0 x 50	05792-61
1.0 x 150	05793-51
2.0 x 30	05878-51
2.0 x 50	05794-41
2.0 x 100	05470-71
2.0 x 150	34449-71
2.0 x 250	05795-31
3.0 x 100	05796-21
3.0 x 150	05797-11
3.0 x 250	05798-01
4.6 x 30	05879-41
4.6 x 50	34451-21

Column Size I.D. x Length (mm)	Product Number
4.6 x 100	05799-91
4.6 x 150	02486-71
4.6 x 250	02485-81
6.0 x 150	34419-61
6.0 x 250	05800-41
10 x 50	05801-31
10 x 150	34466-41
10 x 250	34376-21
20 x 150	34476-11
20 x 250	34373-51
28 x 250	34456-71

COSMOSIL $5C_{18}$-PAQ Guard Column

Column Size I.D. x Length (mm)	Product Number
4.6 x 10	02484-91
10 x 20	34457-61
20 x 20	05803-11
20 x 50	05804-01
28 x 50	34455-81

● Preparative Column (Particle Size: 15 µm)

COSMOSIL $15C_{18}$-PAQ Packed Column

Column Size I.D. x Length (mm)	Product Number
28 x 250	05888-21
50 x 250	05890-71
50 x 500	05891-61

COSMOSIL $15C_{18}$-PAQ Guard Column

Column Size I.D. x Length (mm)	Product Number
28 x 50	05887-31
50 x 50	05889-11

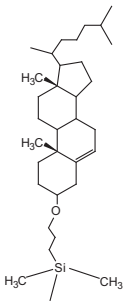
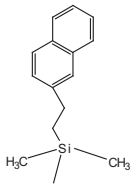
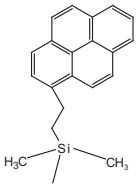
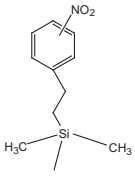
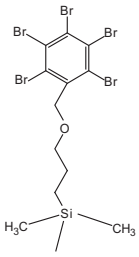
For $15C_{18}$-PAQ, please refer to page 33.
For flow rate and equipment of semi-micro columns, or scale up to preparative columns, please refer to page 189.
For $5C_{18}$-P, please refer to page 59.

(2) Special Columns

Introduction

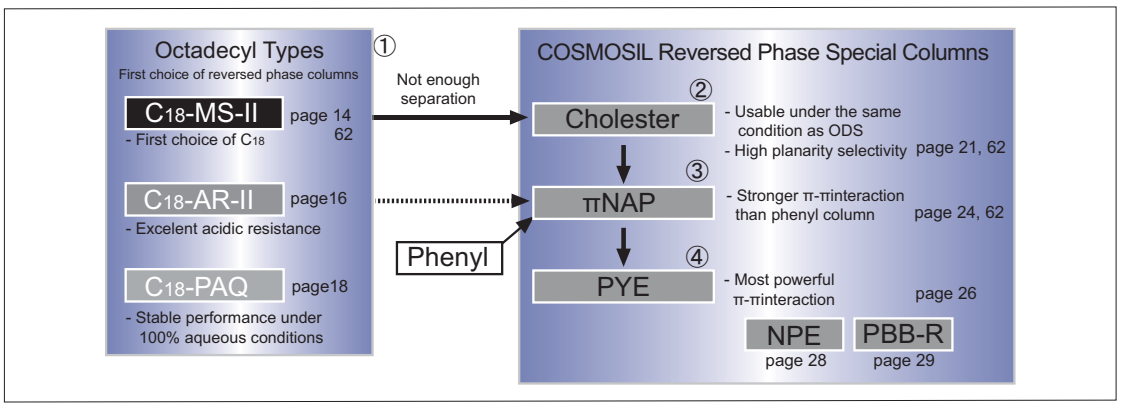
Reversed phase HPLC columns have been widely used because of their superior resolution, high theoretical plate number and ease of use. Since hydrophobic interaction is the dominant separation mechanism in reversed phase chromatography, conventional stationary phases such as C_{18} and C_8 do not offer optimum selectivity for compounds with similar hydrophobicity. COSMOSIL offers a broad selection of columns with unique stationary phases for separation of these difficult analytes. These columns offer improved separation of structurally similar compounds that are difficult to analyze with a C_{18} type column.

Specifications

Packing Material	Cholester	πNAP	PYE	NPE	PBB-R
Silica Gel	High Purity Porous Spherical Silica				
Average Particle Size	5 μm*		5 μm		
Average Pore Size	approx. 120 Å				
Specific Surface Area	approx. 300 m^2/g				
Bonded Phase Stracture					
Bonded Phase	Cholesteryl Group	Naphtylethyl Group	Pyrenylethyl Group	Nitrophenylethyl Group	Pentabromobezyl Group
Bonding Type	Monomeric				
Main Interaction	• Hydrophobic Interaction • Molecular Shape Selectivity	• Hydrophobic Interaction • π-π Interaction	• Hydrophobic Interaction • π-π Interaction • Dispersion Force • Charge-tansfer Interaction	• Hydrophobic Interaction • π-π Interaction • Dipole-dipole Interaction	• Hydrophobic Interaction • Dispersion force
End-capping Treatment	Near-perfect Treatment				
Carbon Load	approx. 20%	approx. 11%	approx. 18%	approx. 9%	approx. 8%
Features	• Usable under condition the same as C_{18} • High molecular sharp selectivity	• Stronger π-π interaction than phenyl column	• Strongest π-π interaction	• Dipole-dipole interaction	• Dispersion force interaction

*For 2.5Cholester, 2.5πNAP (2.5 μm), please refer to page 62.
For more information on interaction, refere to interaction, please refer to Technical Information 7, Selectivity of packing materials in reversed phase liquid chromatography at page 201.

Column Slection Guide



The number in Selection Guide: The order of selection column

COSMOSIL Cholester

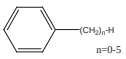
- Cholesterol bonded stationary phase
- Usable under condition the same as C_{18}
- Increased stereoselectivity and improved resolution for geometric isomers
- For separation of natural compounds

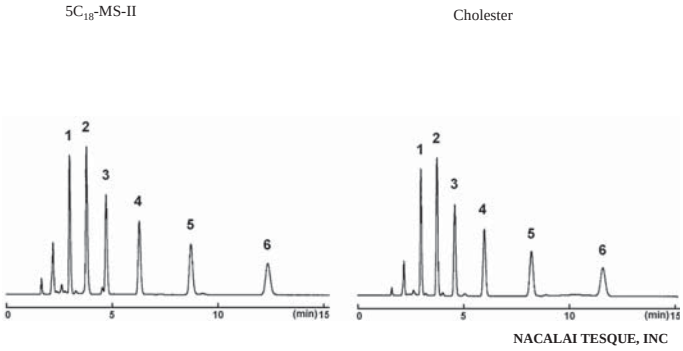
Hydrophobic Interaction

Figure shows the comparison of hydrophobic interactions with competitive C_{18} columns. Cholester provides the same hydrophobicity as alkyl group bonded types (C_{18}, C_{30}). It is not necessary to change the analytical conditions when replacing C_{18} or C_{30} columns with Cholester.

Comparison of Hydrophobic Interaction

Column:
Column size: 4.6mmI.D.-150mm
Mobile phase: Methanol/H_2O = 80/20
Flow rate: 1.0 ml/min
Temperature: 30°C
Detection: UV254nm
Sample:
1; Benzene (1.67 μg)
2; Toluene (1.67 μg)
3; Ethylbenzene (1.67 μg)
4; *n*-Propylbenzene (1.67 μg)
5; *n*-Butylbenzene (1.67 μg)
6; Amylbenzene (1.67 μg)





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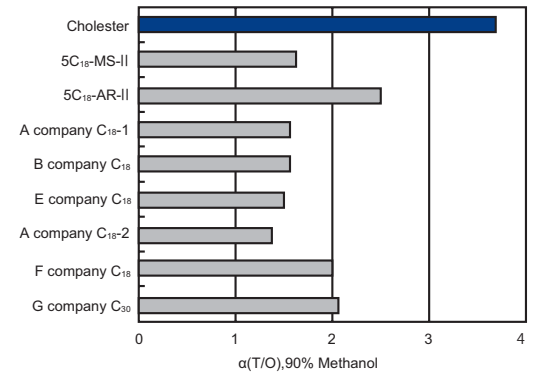
Molecular Shape Selectivity

The stationary phase of Cholester has very rigid structures and can distinguish different molecular shapes. Cholester offers improved separation for structurally similar compounds that are difficult to analyze with alkyl group bonded materials (C_{18} and C_{30}). As in the following example Cholester retains planar Triphenylene longer than stereoscopic *o*-Terphenyl.

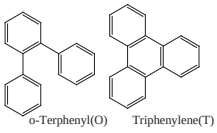


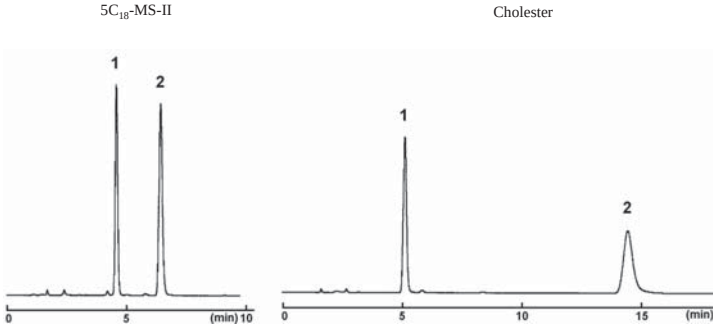
* Particle size: 5 μm(except F company: 3 μm)

Figure. Comparison of molecular sharp selectivity

Comparison of Molecular Shape Selectivity

Column:
Column size: 4.6mmI.D.-150mm
Mobile phase: Methanol/H_2O = 90/10
Flow rate: 1.0 ml/min
Temperature: 30°C
Detection: UV254nm
Sample:
1; *o*-Terphenyl (0.1 μg)
2; Triphenylene (0.01 μg)





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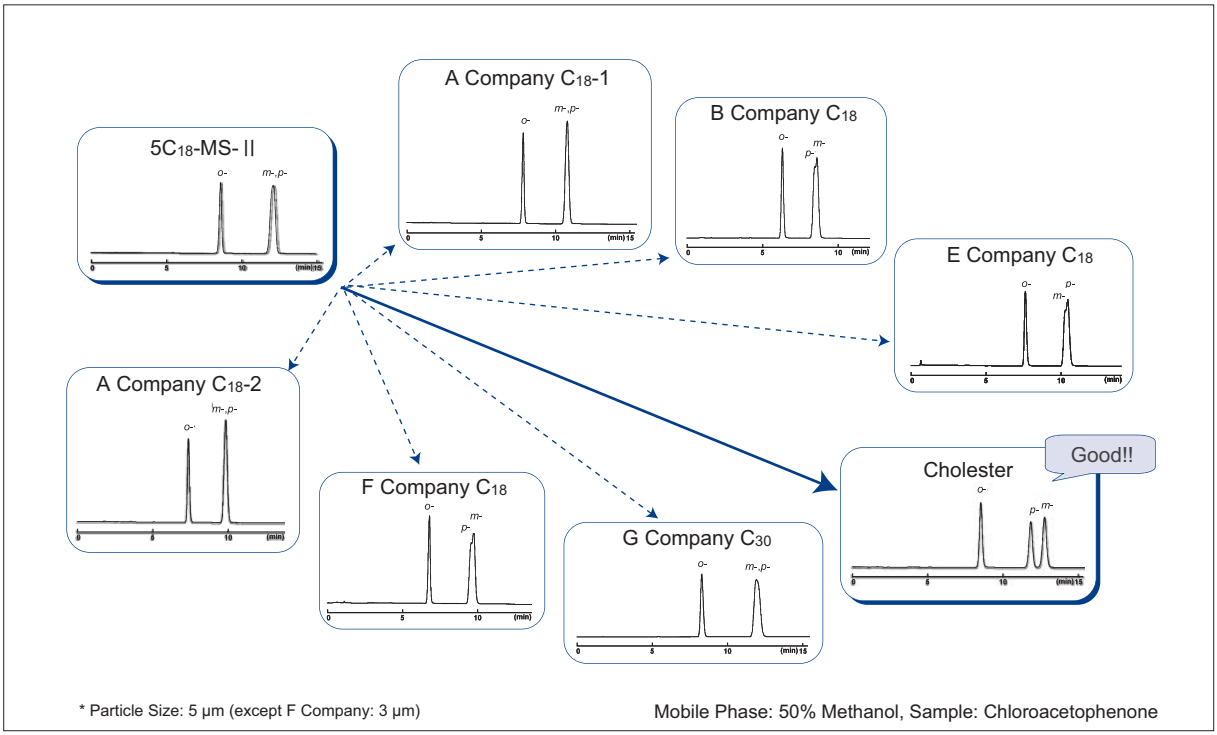
Improvement in Separation

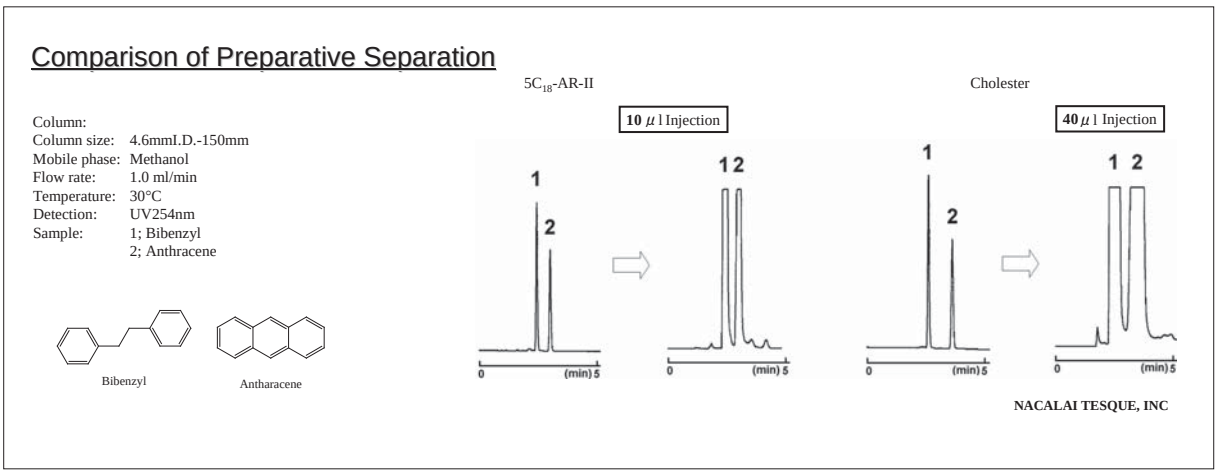
COSMOSIL Cholester provides enhanced selectivity over traditional C_{18} columns and offers greater performance in separating isomers or other closely related compounds. COSMOSIL Cholester is ideal for method development and serves as an excellent alternative to traditional C_{18} columns. The figure below shows analytical data of chloroacetophenone isomers. These isomers are difficult to separate with C_{18} and C_{30}, but they are well resolved by COSMOSIL Cholester.

Comparison with Competitors' C_{18} and C_{30} columns



Efficiency of Preparative Separation

The figure below shows the comparison of efficiency of preparative separation with a C_{18} column. Both columns show good separation. However, sample loading capacity for preparative separations can be affected by a slight difference in separation ability. COSMOSIL Cholester can load 4 times of sample volume compared with C_{18} columns.



Applications

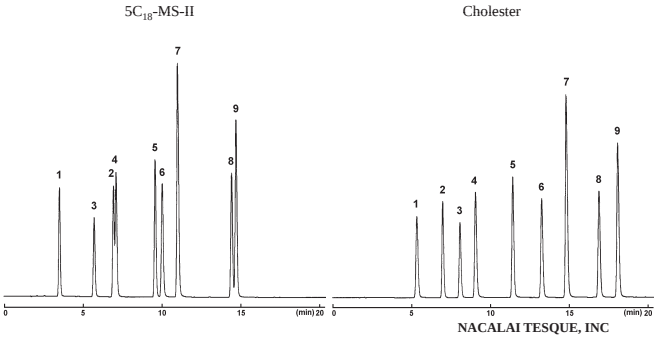
● Catechins

COSMOSIL Application Data

Column:
Column size: 4.6mmI.D.-150mm
Mobile phase: A: Acetonitrile/ 20mmol/l Phosphate buffer(pH2.5) = 10/90
B: Acetonitrile/ 20mmol/l Phosphate buffer(pH2.5) = 30/70
B conc. 0→100% 20min Linear gradient
Flow rate: 1.0 ml/min
Temperature: 30°C
Detection: UV280nm
Sample:

1; (-) - Gallocatechin〔(-) - GC〕	(0.80mg/ml)
2; Caffeine	(0.08mg/ml)
3; (-) - Epigallocatechin〔(-) - EGC〕	(0.80mg/ml)
4; (-) - Catechin〔(-) - C〕	(0.40mg/ml)
5; (-) - Epicatechin〔(-) - EC〕	(0.40mg/ml)
6; (-) - Epigallocatechin Gallate〔(-) - EGCg〕	(0.20mg/ml)
7; (-) - Gallocatechin Gallate〔(-) - GCg〕	(0.40mg/ml)
8; (-) - Epicatechin Gallate〔(-) - ECg〕	(0.20mg/ml)
9; (-) - Catechin Gallate〔(-) - Cg〕	(0.20mg/ml)

Injection Vol. 1.0µl



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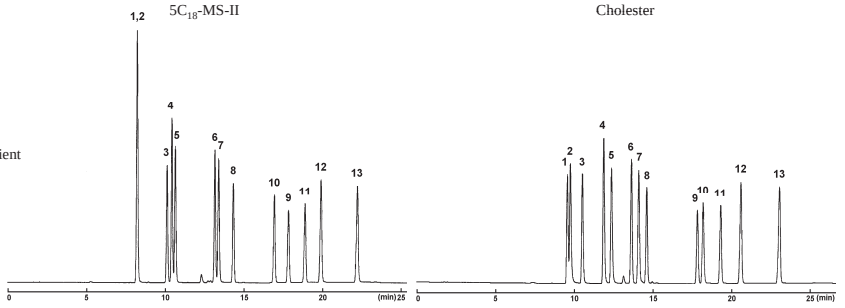
● Flavones

COSMOSIL Application Data

Column:
Column size: 4.6mmI.D.-150mm
Mobile phase: A: Acetonitrile/ 20mmol/l Phosphate buffer(pH2.5) = 20/80
B: Acetonitrile/ 20mmol/l Phosphate buffer(pH2.5) = 70/30
B conc. 0→100% 20min Linear gradient
Flow rate: 1.0 ml/min
Temperature: 30°C
Detection: UV280nm
Sample:

1; Fisetin	(0.25µg)		
2; Myricetin	(0.20µg)		
3; 7,8-Dihydroxyflavone	(0.05µg)		
4; Luteolin	(0.20µg)		
5; Quercetin	(0.20µg)		
6; 7-Hydroxyflavone	(0.10µg)	10; Chrysin	(0.05µg)
7; Baicalein	(0.05µg)	11; 6-Methoxyflavone	(0.05µg)
8; 6-Hydroxyflavone	(0.05µg)	12; 3-Hydroxyflavone	(0.25µg)
9; Flavone	(0.05µg)	13; 5-Hydroxyflavone	(0.05µg)



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Ordering Information

● Analytical / Preparative Column (Particle Size: 5 µm)

COSMOSIL Cholester Packed Column

Column Size I.D. x Length (mm)	Product Number
1.0 x 150	05968-71
1.0 x 250	05969-61
2.0 x 30	08565-51
2.0 x 50	06352-91
2.0 x 100	06948-01
2.0 x 150	05971-11
2.0 x 250	05972-01
3.0 x 150	05973-91
3.0 x 250	05974-81

Column Size I.D. x Length (mm)	Product Number
4.6 x 150	05976-61
4.6 x 150 3 lots set*	07970-03
4.6 x 250	05977-51
10 x 150	08011-91
10 x 250	05979-31
20 x 150	06088-71
20 x 250	05982-71
28 x 250	05985-41

*For 4.6 x 150 3 lots set, please refer to page 11.

COSMOSIL Cholester Guard Column

Column Size I.D. x Length (mm)	Product Number
4.6 x 10	05975-71
10 x 20	05978-41
20 x 20	05980-91
20 x 50	05981-81
28 x 50	05983-61

For more information on 2.5Cholester (2.5 µm), please refer to page 62.

COSMOSIL πNAP

- Naphthalene bonded stationary phase
- Enhanced π-π interactions
- Improved selectivity for structural isomers

Comparison of π-π interactions

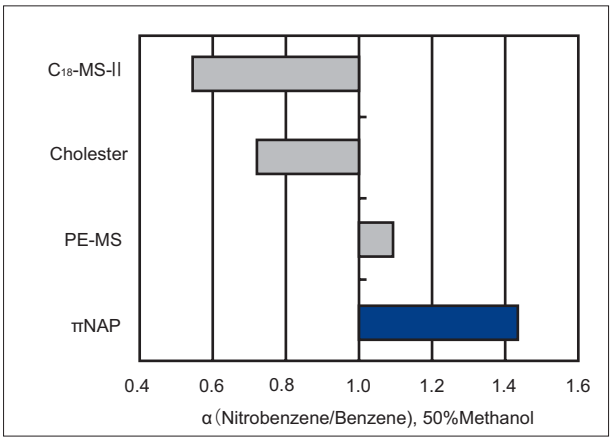


Figure. Comparison of π-π interaction

COSMOSIL πNAP shows stronger π-π interactions than phenyl columns. Its two fused aromatic rings retain nitrobenzene with more π electrons stronger than phenyl columns.

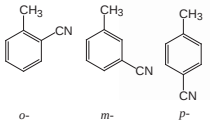
Applications

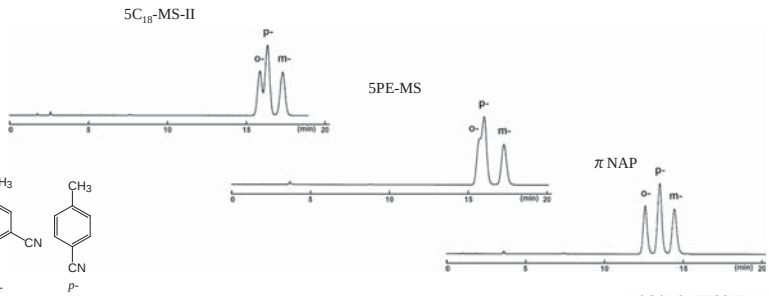
- Tolunitriles

COSMOSIL Application Data

Column:
Column size: 4.6mmI.D.-150mm
Mobile phase: $5C_{18}$-MS-II Methanol/ H_2O = 40/60
5PE-MS Methanol/ H_2O = 40/60
π NAP Methanol/ H_2O = 50/50
Flow rate: 1.0 ml/min
Temperature: 30°C
Detection: UV254nm

Sample: *o*-Tolunitrile (2.0 μg)
m-Tolunitrile (2.0 μg)
p-Tolunitrile (1.0 μg)





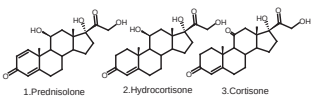
NACALAI TESQUE, INC
AP-1022

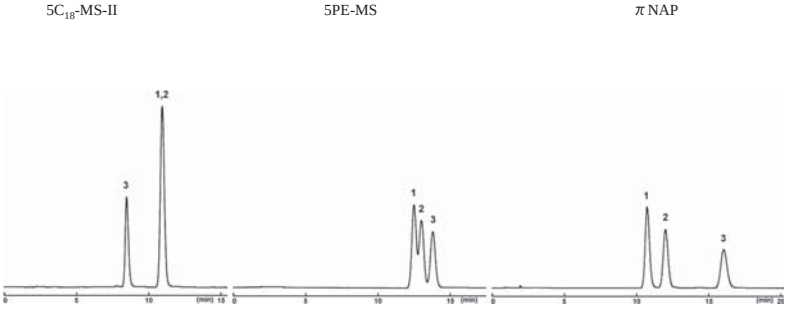
- Adrenal Cortical Hormones

COSMOSIL Application Data

Column:
Column size: 4.6mmI.D.-150mm
Mobile phase: $5C_{18}$-MS-II Methanol/ H_2O = 50/50
5PE-MS Methanol/ H_2O = 50/50
π NAP Methanol/ H_2O = 60/40
Flow rate: 1.0 ml/min
Temperature: 30°C
Detection: UV254nm

Sample: 1; Prednisolone (0.33 μg)
2; Hydrocortisone (0.33 μg)
3; Cortisone (0.33 μg)





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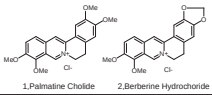
Applications

● Berberines

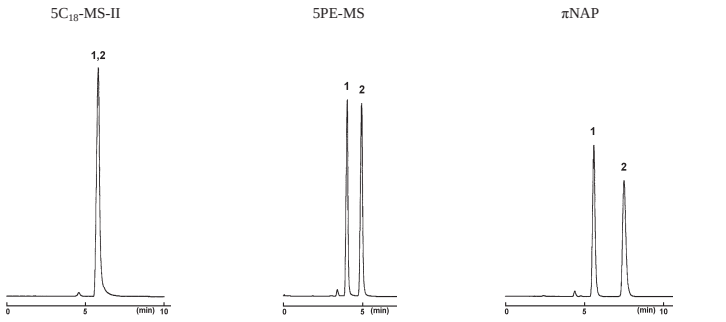
COSMOSIL Application Data

Column:
Column size: 4.6mmI.D.-150mm
Mobile phase: Methanol/ 20mmol/l Phosphate buffer(pH2.5)
$5C_{18}$-MS-II = 40/60
5PE-MS = 60/40
πNAP = 80/20
Flow rate: 1.0 ml/min
Temperature: 30°C
Detection: UV254nm

Sample: 1; Palmatine (0.2μg)
2; Berberine (0.2μg)



1,Palmatine Cholide 2,Berberine Hydrochoride



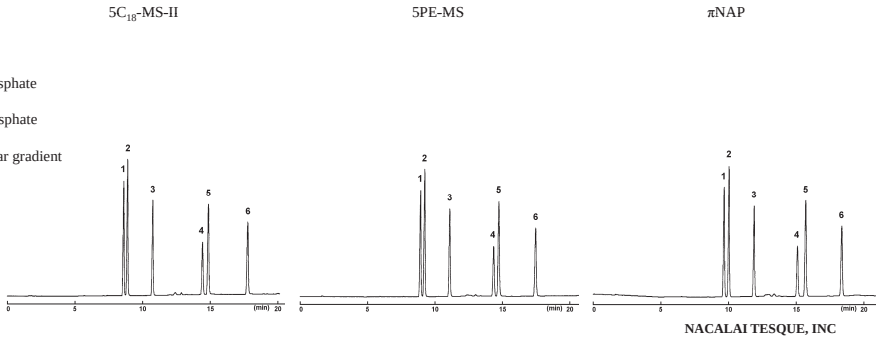
AP-1024

● Isoflavones

COSMOSIL Application Data

Column:
Column size: 4.6mmI.D.-150mm
Mobile phase: A: Acetonitrile/ 20mmol/l Phosphate buffer(pH2.5) = 10/90
B: Acetonitrile/ 20mmol/l Phosphate buffer(pH2.5) = 50/50
B conc. 0→100% 20min Linear gradient
Flow rate: 1.0 ml/min
Temperature: 30°C
Detection: UV260nm

Sample: 1; Daidzin (0.05μg)
2; Glycitin (0.05μg)
3; Genistin (0.05μg)
4; Daidzein (0.05μg)
5; Glycitein (0.075μg)
6; Genistein (0.025μg)



AP-1025

Ordering Information

● Analytical / Preparative Column (Particle Size: 5 μm)

COSMOSIL πNAP Packed Column

Column Size I.D. x Length (mm)	Product Number
1.0 x 150	08076-61
1.0 x 250	08077-51
2.0 x 30	08566-41
2.0 x 50	08567-31
2.0 x 100	08299-51
2.0 x 150	08078-41
2.0 x 250	08079-31
3.0 x 150	08080-91

Column Size I.D. x Length (mm)	Product Number
3.0 x 250	08081-81
4.6 x 150	08085-41
4.6 x 250	08086-31
10 x 150	08088-11
10 x 250	08089-01
20 x 150	08092-41
20 x 250	08093-31
28 x 250	08095-11

COSMOSIL πNAP Guard Column

Column Size I.D. x Length (mm)	Product Number
4.6 x 10	08082-71
10 x 20	08087-21
20 x 20	08090-61
20 x 50	08091-51
28 x 50	08094-21

For more information on 2.5πNAP (2.5 μm), please refer to page 62.

COSMOSIL PYE

- Pyrenylethyl group bonded stationary phase
- Separation with high molecular shape selectivity or π-π interactions
- Excellent separation for structural isomers

Comparison of π-π interaction

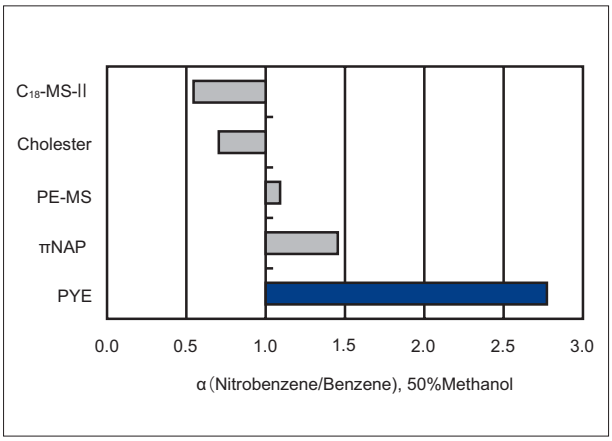
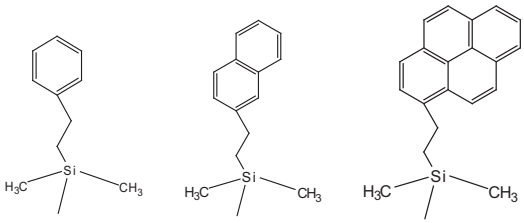


Figure. Comparison of π-π interactions

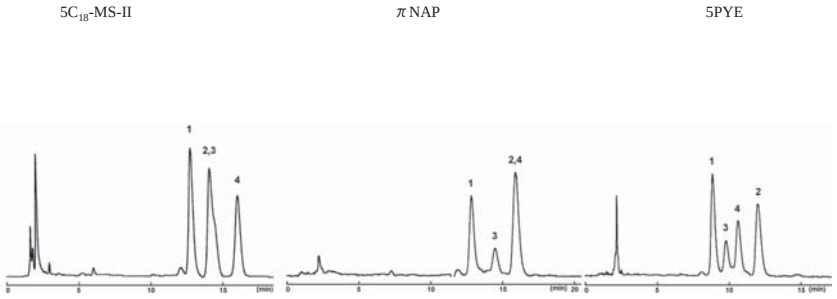
COSMOSIL PYE provides much stronger π-π interactions than πNAP on page 24.



Applications

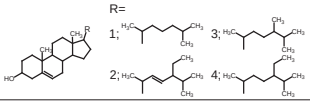
- Sterols

COSMOSIL Application Data



Column:
Column size: 4.6mmI.D.-150mm
Mobile phase: $5C_{18}$-MS-II Methanol/ H_2O = 98/2
π NAP Methanol/ H_2O = 90/10
5PYE Methanol/ H_2O = 95/5
Flow rate: 1.0 ml/min
Temperature: 30°C
Detection: UV210nm

Sample: 1; Cholesterol (5.6 μg)
2; Stigmasterol (5.9 μg)
3; Campesterol
4; Sitosterol



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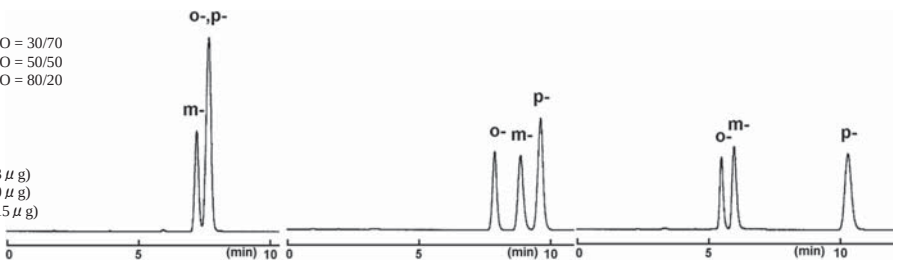
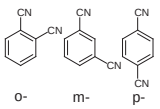
- Phthalonitriles

COSMOSIL Application Data



Column:
Column size: 4.6mmI.D.-150mm
Mobile phase: $5C_{18}$-MS-II Methanol/ H_2O = 30/70
π NAP Methanol/ H_2O = 50/50
5PYE Methanol/ H_2O = 80/20
Flow rate: 1.0 ml/min
Temperature: 30°C
Detection: UV254nm

Sample: *o*-; Phthalonitrile (0.3 μg)
m-; Isophthalonitrile (3.0 μg)
p-; Terephthalonitrile (0.15 μg)



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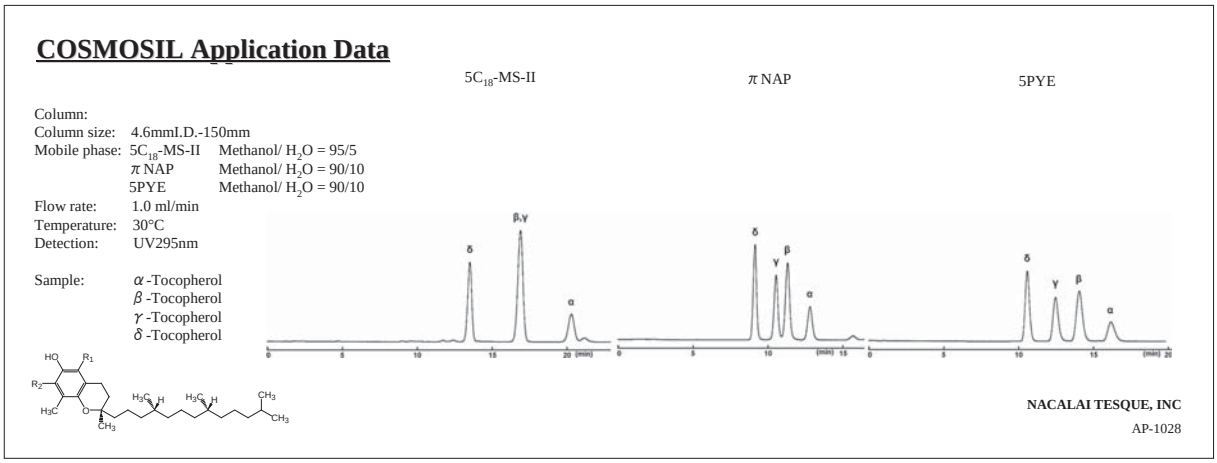
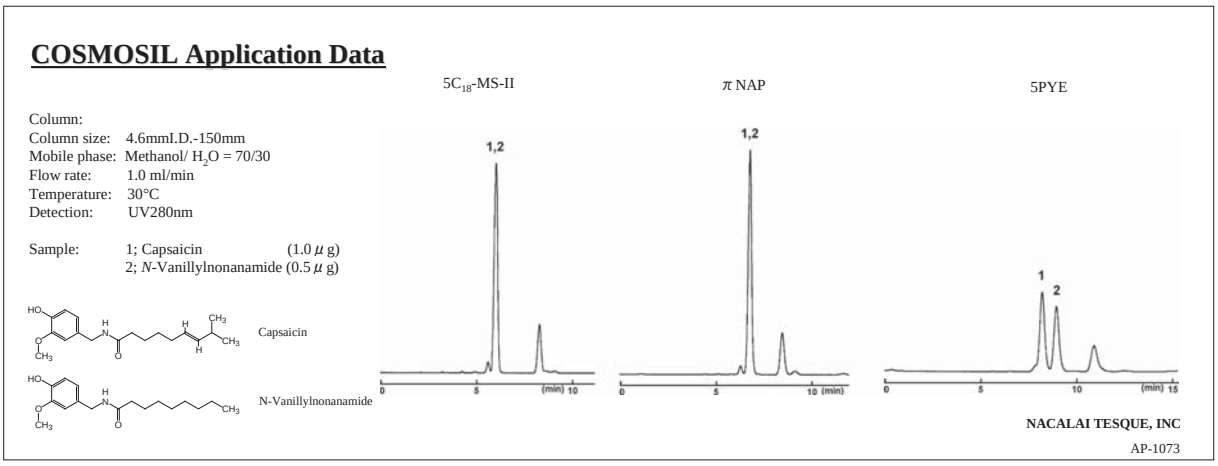
Applications

● Tocopherols



● Capsaicins



Attention

1. Methanol is recommended as a mobile phase for COSMOSIL PYE column. Acetonitrile is not recommended because it has many π electrons and interferes π-π interactions between a sample and the stationary phase.
2. The stationary phase of COSMOSIL PYE, pyrenylethyl group, has a large UV absorption. When the stationary phase detaches from silica gel and elutes, even a slight quantity can be detected and causes baseline noise. In such a case, wash the column with tetrahydrofuran. Detachment of a small amount of the stationary phase does not deteriorate a column's separation ability.
3. COSMOSIL PYE column is not suitable for gradient analysis.

Ordering Information

● Analytical / Preparative Column (Particle Size: 5 μm)

COSMOSIL 5PYE Packed Column

Column Size I.D. x Length (mm)	Product Number
1.0 x 150	02851-71
2.0 x 150	38042-61
2.0 x 250	34450-31

Column Size I.D. x Length (mm)	Product Number
4.6 x 150	37837-91
4.6 x 250	37989-11
10 x 250	37996-11
20 x 250	38044-41

COSMOSIL 5PYE Guard Column

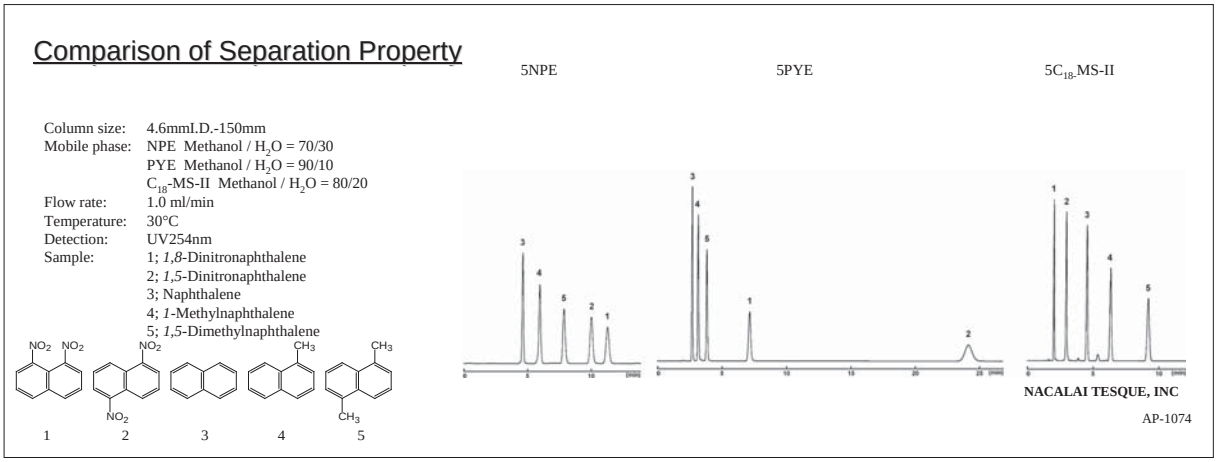
Column Size I.D. x Length (mm)	Product Number
4.6 x 10	37903-11
10 x 20	38041-71
20 x 20	05867-91
20 x 50	34475-21

COSMOSIL NPE

- Nitrophenylethyl group bonded stationary phase
- Separation with dipole-dipole and π-π interactions
- Excellent separation for structural isomers

Selectivity for Dipole-dipole Interactions

COSMOSIL NPE strongly retains *1,8*-dinitronaphthalene because of the strong dipole formed by the two nitro groups positioned on the same side of naphthalene.



Attention

1. Methanol is recommended as a mobile phase for COSMOSIL NPE column. Acetonitrile is not recommended because it has many π electrons and interferes π-π interactions between a sample and the stationary phase.
2. The stationary phase of COSMOSIL NPE, nitrophenyl group, has a large UV absorption. When the stationary phase detaches from silica gel and elutes, even a slight quantity can be detected and causes baseline noise. In such a case, wash the column with tetrahydrofuran. Detachment of a small amount of the stationary phase does not deteriorate a column's separation ability.
3. COSMOSIL NPE column is not suitable for gradient analysis.

Ordering Information

● Analytical / Preparative Column (Particle Size: 5 µm)

COSMOSIL 5NPE Packed Column

Column Size I.D. x Length (mm)	Product Number
1.0 x 150	05897-01
2.0 x 150	34328-51
2.0 x 250	34379-91
4.6 x 150	37902-21
4.6 x 250	37990-71
10 x 250	05469-11
20 x 250	38046-21

COSMOSIL 5NPE Guard Column

Column Size I.D. x Length (mm)	Product Number
4.6 x 10	37904-01
10 x 20	38045-31
20 x 20	05868-81
20 x 50	05869-71

COSMOSIL PBB-R

- Pentabromobenzyl bonded stationary phase
- Separation with dispersion force interaction
- Distinguish various surfactants' polyethylene glycol chain length
- For separation of halogen exchange reaction products or aromatic compounds

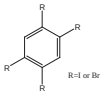
Applications

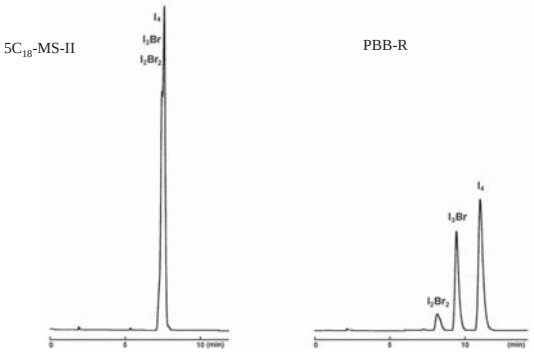
● Halogen Exchange Reaction Products

COSMOSIL PBB-R strongly retains iodine atom which has a large dispersion force, than bromine atom. So it can separate halogen exchange reaction products that are difficult to analyze with C_{18} column.

COSMOSIL Application Data

Column:
Column size: 4.6mmI.D.-150mm
Mobile phase: $5C_{18}$-MS-II Methanol/ H_2O = 90/10
PBB-R Methanol
Flow rate: 1.0 ml/min
Temperature: 30°C
Detection: UV254nm
Sample:





Sample courtesy of Dr.H.Yamamoto,RIKEN

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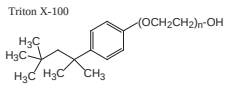
● Surfactant Agents

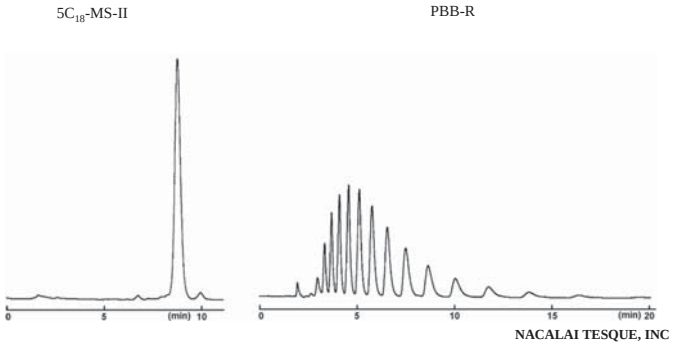
C_{18} column can not separate Triton X-100 mixturem, because (-OCH_2CH_2-) group has little hydrophobicity. However, COSMOSIL PBB-R can separate them because it distinguishes difference in the dispersion force, which depends on its molecular weight.

COSMOSIL Application Data

Column:
Column size: 4.6mmI.D.-150mm
Mobile phase: $5C_{18}$-MS-II Methanol/ H_2O = 80/20
PBB-R Methanol
Flow rate: 1.0 ml/min
Temperature: 30°C
Detection: UV254nm
Sample: Triton X-100 ($5C_{18}$-MS-II: 50 μg)
(5PBB-R : 100 μg)

Triton X-100





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Ordering Information

● Analytical / Preparative Column (Particle Size: 5 μm)

COSMOSIL 5PBB-R Packed Column

Column Size I.D. x Length (mm)	Product Number
1.0 x 150	05899-81
2.0 x 150	05900-31
2.0 x 250	05904-91

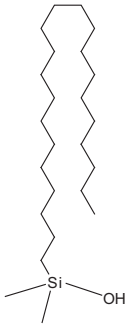
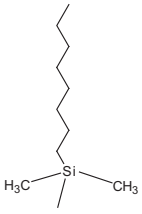
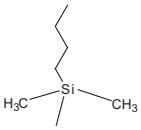
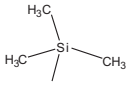
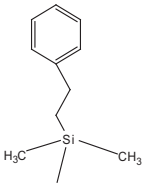
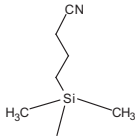
Column Size I.D. x Length (mm)	Product Number
4.6 x 150	05697-21
4.6 x 250	05698-11
10 x 250	05699-01
20 x 250	05700-51

COSMOSIL 5PBB-R Guard Column

Column Size I.D. x Length (mm)	Product Number
4.6 x 10	05704-11
10 x 20	05721-81
20 x 20	05911-91
20 x 50	05722-71

(3) Other Reversed Phase HPLC Columns

Specifications

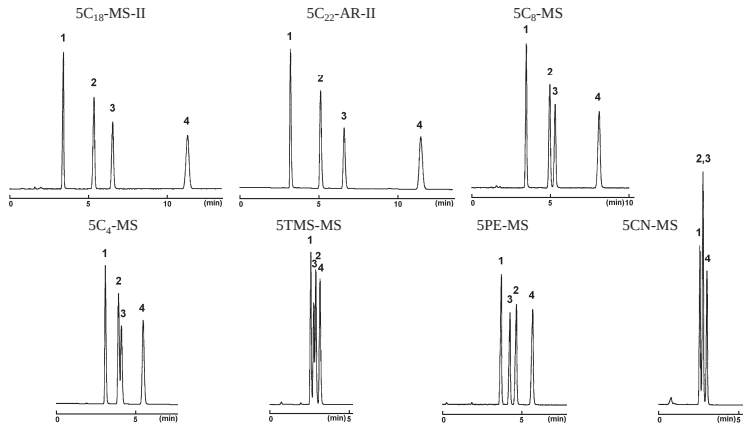
Packing Material	C_{22}-AR-II	C_8-MS	C_4-MS	TMS-MS	PE-MS	CN-MS
Silica Gel	High Purity Porous Spherical Silica					
Average Particle Size	5 µm					
Average Pore Size	approx. 120 Å					
Specific Surface Area	approx. 300 m^2/g					
Bonded Phase Stracture						
Bonded Phase	Dococyl Group	Octyl Group	Butyl Group	Trimethyl Group	Phenylethyl Group	Cyanopropyl Group
Bonding Type	Polymeric	Monomeric				
Main Interaction	Hydrophobic Interaction				Hydrophobic Interaction π-π Interaction	
End-capping Treatment	Near-perfect Treatment					
Carbon Load	approx. 19%	approx. 10%	approx. 7%	approx. 5%	approx. 10%	approx. 7%

Different of Separation Characteristic

COSMOSIL Application Data

Column:
Column size: 4.6mmI.D.-150mm
Mobile phase: Methanol/ H_2O = 60/40
Flow rate: 1.0 ml/min
Temperature: 30°C
Detection: UV254nm

Sample:
1; Acetophenone (0.05µg)
2; Methyl Benzoate (0.5µg)
3; Benzene (2.0µg)
4; Toluene (2.0µg)

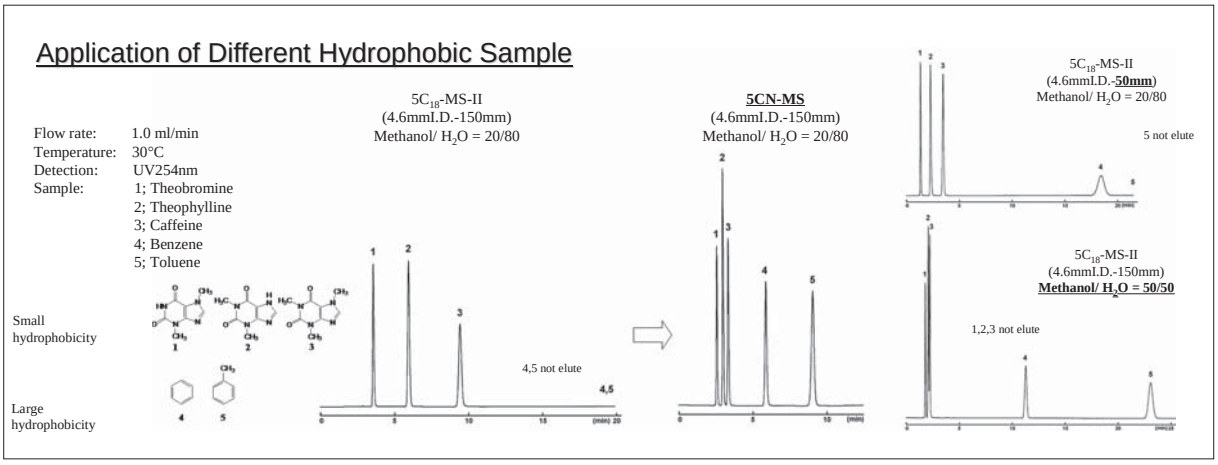


COSMOSIL CN-MS

- Cyanopropyl group bonded stationary phase
- Enables separation of different hydrophobic samples without using gradient
- For separation of natural compounds with different hydrophobicity

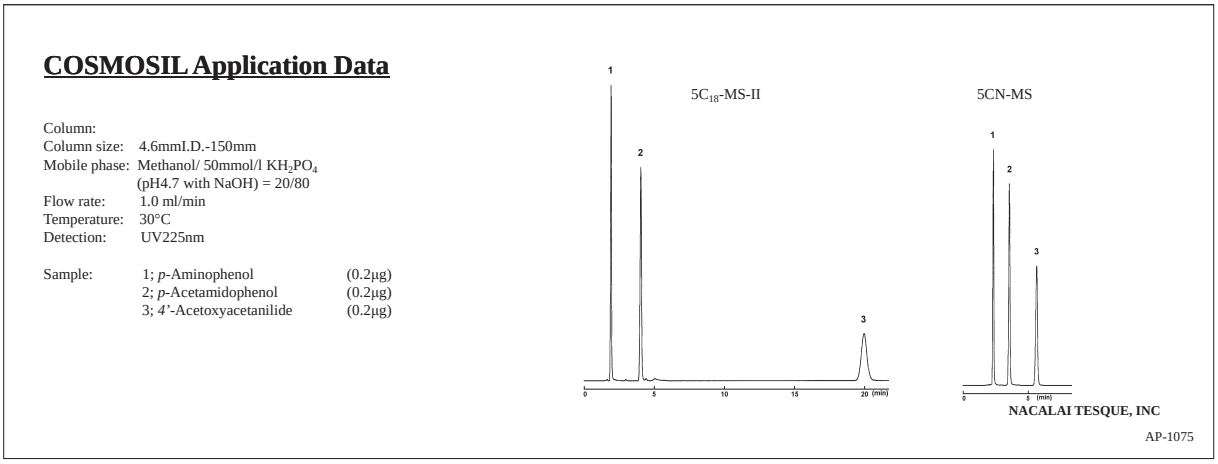
Rapid Analysis

Gradient elution is commonly used for the samples containing both polar and non-polar compounds. However, gradient elution may cause reproducibility problem depending on the gradient mixer and pump, and need an equilibration time for each analysis. COSMOSIL 5CN-MS offers rapid analysis and great reproducibility using isocratic elution mode.



Applications

●Acetoaminophen



Ordering Information

● Analytical / Preparative Column (Particle Size: 5 µm)

COSMOSIL 5CN-MS Packed Column

Column Size I.D. x Length (mm)	Product Number
4.6 x 50	38233-61
4.6 x 100	38234-51
4.6 x 150	38235-41
4.6 x 250	38236-31

Column Size I.D. x Length (mm)	Product Number
6.0 x 150	38237-21
6.0 x 250	38238-11
10 x 250	38239-01
20 x 250	38240-61

COSMOSIL 5CN-MS Guard Column

Column Size I.D. x Length (mm)	Product Number
4.6 x 10	38231-81
10 x 20	38232-71

COSMOSIL C_{22}-AR-II, C_8-MS, C_4-MS, TMS-MS, PE-MS

Ordering Information

● Analytical / Preparative Column (Particle Size: 5 µm)

COSMOSIL 5C_{22}-AR-II Packed Column

Column Size I.D. x Length (mm)	Product Number
4.6 x 50	05848-41
4.6 x 100	05849-31
4.6 x 150	04598-51
4.6 x 250	04599-41

Column Size I.D. x Length (mm)	Product Number
6.0 x 150	05850-91
6.0 x 250	05851-81
10 x 250	04969-91
20 x 250	05183-41

COSMOSIL 5C_{22}-AR-II Guard Column

Column Size I.D. x Length (mm)	Product Number
4.6 x 10	04881-21
10 x 20	05554-81

COSMOSIL 5C_8-MS Packed Column

Column Size I.D. x Length (mm)	Product Number
4.6 x 50	38153-11
4.6 x 100	38154-01
4.6 x 150	38155-91
4.6 x 250	38156-81

Column Size I.D. x Length (mm)	Product Number
6.0 x 150	38157-71
6.0 x 250	38158-61
10 x 250	38159-51
20 x 250	38160-11

COSMOSIL 5C_8-MS Guard Column

Column Size I.D. x Length (mm)	Product Number
4.6 x 10	38151-31
10 x 20	38152-21

COSMOSIL 5C_4-MS Packed Column

Column Size I.D. x Length (mm)	Product Number
4.6 x 50	38163-81
4.6 x 100	38164-71
4.6 x 150	38165-61
4.6 x 250	38166-51

Column Size I.D. x Length (mm)	Product Number
6.0 x 150	38167-41
6.0 x 250	38168-31
10 x 250	38169-21
20 x 250	38170-81

COSMOSIL 5C_4-MS Guard Column

Column Size I.D. x Length (mm)	Product Number
4.6 x 10	38161-01
10 x 20	38162-91

COSMOSIL 5TMS-MS Packed Column

Column Size I.D. x Length (mm)	Product Number
4.6 x 50	38173-51
4.6 x 100	38174-41
4.6 x 150	38175-31
4.6 x 250	38176-21

Column Size I.D. x Length (mm)	Product Number
6.0 x 150	38177-11
6.0 x 250	38178-01
10 x 250	38179-91
20 x 250	38180-51

COSMOSIL 5TMS-MS Guard Column

Column Size I.D. x Length (mm)	Product Number
4.6 x 10	38171-71
10 x 20	38172-61

COSMOSIL 5PE-MS Packed Column

Column Size I.D. x Length (mm)	Product Number
4.6 x 50	38183-21
4.6 x 100	38184-11
4.6 x 150	38185-01
4.6 x 250	38186-91

Column Size I.D. x Length (mm)	Product Number
6.0 x 150	38187-81
6.0 x 250	38188-71
10 x 250	38189-61
20 x 250	38190-21

COSMOSIL 5PE-MS Guard Column

Column Size I.D. x Length (mm)	Product Number
4.6 x 10	38181-41
10 x 20	38182-31

COSMOSIL 15C_{18}-MS-II, 15C_{18}-AR-II, 15C_{18}-PAQ

- 2 sizes of inner diameter (28 mm and 50 mm)
- 15 µm silica gel

Specifications

Packing Material	15C_{18}-MS-II	15C_{18}-AR-II	15C_{18}-PAQ
Silica Gel	High Purity Porous Spherical Silica		
Average Particle Size	15 µm		
Average Pore Size	approx. 120 Å		
Specific Surface Area	approx. 300 m^2/g		
Bonded Phase Stracture	Please refer to page 12		
Bonded Phase	Octadecyl Group		
Bonding Type	Monomeric	Polymeric	
Main Interaction	Hydrophobic Interaction		
End-capping Treatment	Near-perfect Treatment		
Usable pH Range	2~10	1.5~7.5	2~7.5
Carbon Load	approx. 16%	approx. 17%	approx. 11%
Features	• Multi-purpose C_{18} Column • Suitable for basic compounds.	• Features strong acid resistance. • Suitable for acid compounds and peptides.	• Suitable for hydrophilic compounds. • Compatible with 100% water based mobile phase.

Applications

- Preparative separation of carbazole using 50 mm I.D. column

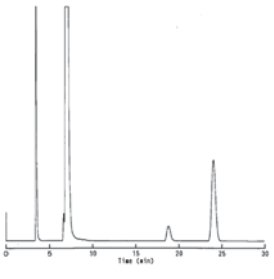
Carbazole is extracted from anthracene oil (coal tar) and required high purity because it is often used for analytical applications. Following is the preparative separation of carbazol using a 50 mm I.D. COSMOSIL 15C_{18}-AR-II.

To Preparative Separation

Analytical

(Analytical)
Column: 5C_{18}-AR-II
Column size: 4.6mmI.D.-150mm
Flow rate: 0.5 ml/min
Temperature: room temperature
Detection: UV254nm

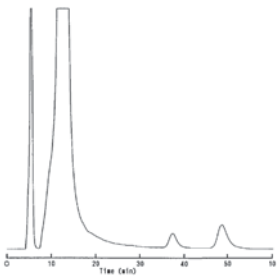
Sample: Carbazole (50mg/ml)
Injection Vol. 0.5 µl (25 µg)



Preparative

(Preparative)
Column: 15C_{18}-AR-II
Column size: 50mmI.D.-250mm
Flow rate: 60ml/min
Temperature: room temperature
Detection: UV254nm

Sample: Carbazole (50mg/ml)
Injection Vol. Preparative 3ml (150mg)



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Refer to Technical Information 2, Inner diameter of column (scale down and scale up) at page 189.

Ordering Information

Refer to page 15 for 15C_{18}-MS-II , page 17 for 15C_{18}-AR-II and page 19 for 15C_{18}-PAQ.

6. Normal Phase Chromatography Column

COSMOSIL SL-II

- High purity silica gel (>99.99%) with special treatment
- Suitable for preparative separation

Specifications

Packing Material	SL-II
Silica Gel	High Purity Porous Spherical Silica
Average Particle Size	3, 5, 15 µm
Average Pore Size	approx. 120 Å
Specific Surface Area	approx. 300 m^2/g
Features	• High purity silica gel (>99.99%) with special treatment • Suitable for preparative separation (high resolution than medium-pressure or open chromatography)

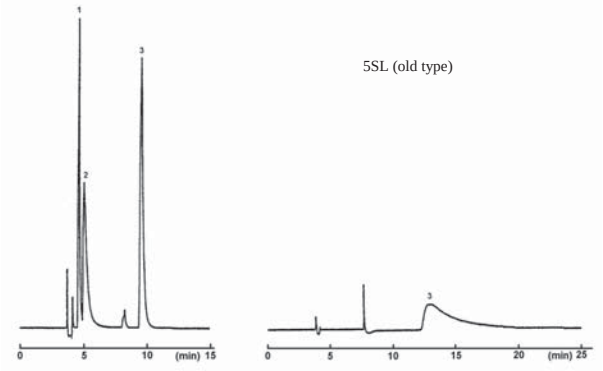
Comparison with Old Type

COSMOSIL SL-II with high purity silica gel offers better peak shape for phenols with simple mobile phase of ethanol or hexane. No acetic acid additives were required, unlike for the old type silica.

Comparison with Old Type

Column:
Column size: 4.6mmI.D.-250mm
Mobile phase: Ethanol/Hexane = 10/90
Flow rate: 1.0 ml/min
Temperature: 30°C
Detection: UV254nm

Sample: 1; Benzoic Acid (4.0 µg)
2; Salicylic Acid (6.0 µg)
3; Salicylamide (4.0 µg)



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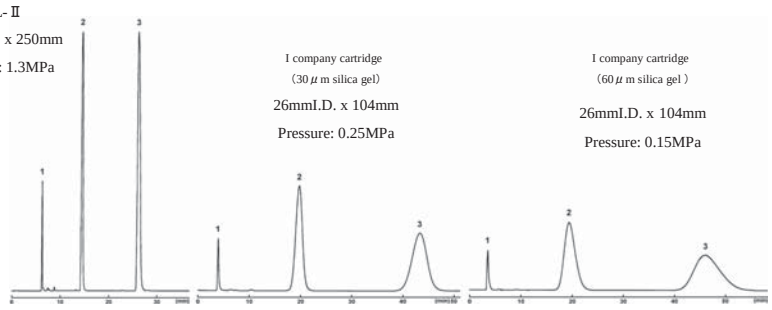
Comparison with Medium-pressure Column

COSMOSIL SL-II offers sharper peak compared with packing materials for medium-pressure liquid chromatography andopen chromatography.

Comparison with Medium-Pressure Columns

Column:
Column size:
Mobile phase: Ethanol/Hexane = 5/95
Flow rate: 10 ml/min
Temperature: room temperature
Detection: UV254nm

Sample: 1; *p*-Xylene (8.3mg)
2; Cinnamyl Alcohol (1.7mg)
3; *p*-Nitrobenzyl Alcohol (4.2mg)



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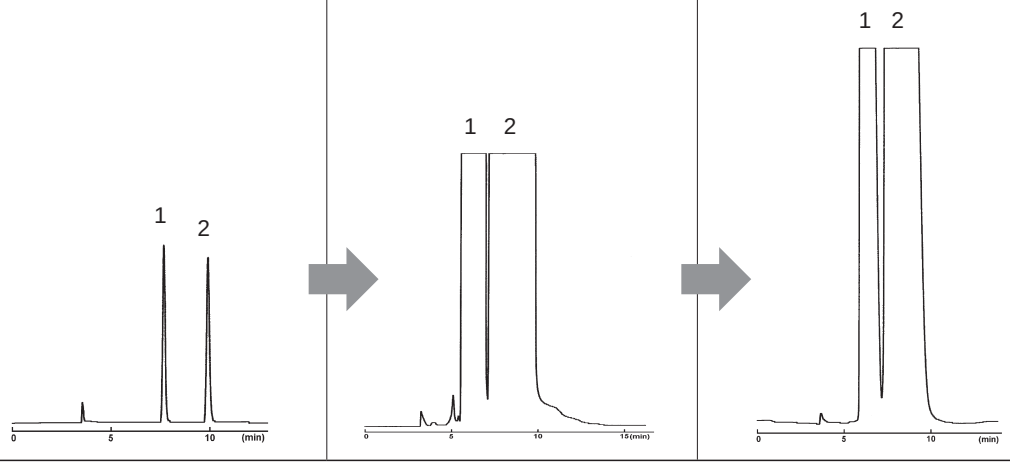
Scaling Up from Analytical to Preparative Separation

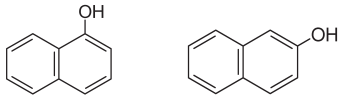
Normal phase chromatography, with non-polar mobile phase and low boiling point is used for preparative separation because solvent removal is generally easier for normal phase chromatography than for reversed chromatography. The followings are how to scale up from analytical (4.6 mm I.D.) to preparative (20.0 mm I.D.) separation.

Step 1	Step 2	Step 3
Mobile phase is optimized by using 4.6 mm I.D. x 250 mm column	Maximum load capacity is optimized by using 4.6 mm I.D. x 250 mm column	Preparative separation by using 20 mm I.D. x 250 mm column
Note As mobile phase, highly soluble solvent for sample is suitable.	Max. injection volume 56 µl (28 mg)	Max. injection volume 1,050 µl (525 mg) (19 times higher than 4.6 mm I.D.) *calculated by cross-section ratio.



Mobile Phase: Ethyl Acetate : Hexane = 10 : 90
Flow Rate: 4.6 mm I.D. : 1 ml/min
20 mm I.D. : 18.9 ml/min
Temperature: Room Temperature
Detection: UV 254 nm

Sample: *1*-Naphtol, *2*-Naphtol



1 2

Please refer to Technical Information 2, Inner diameter of column (scale down and scale up) at page 189.

Ordering Information

● Analytical / Preparative Column (Particle Size: 5 µm)

COSMOSIL 5SL-II Packed Column

Column Size I.D. x Length (mm)	Product Number
4.6 x 50	37999-81
4.6 x 100	38000-01
4.6 x 150	38001-91
4.6 x 250	38002-81

Column Size I.D. x Length (mm)	Product Number
6.0 x 150	38003-71
6.0 x 250	38004-61
10 x 250	38005-51
20 x 250	38006-41
28 x 250	34358-61

COSMOSIL 5SL-II Guard Column

Column Size I.D. x Length (mm)	Product Number
4.6 x 10	37997-01
10 x 20	37998-91
20 x 20	05874-91
20 x 50	05875-81
28 x 50	34359-51

● Preparative Column (Particle Size : 15 µm)

COSMOSIL 15SL-II Packed Column

Column Size I.D. x Length (mm)	Product Number
28 x 250	05893-41
50 x 250	05895-21
50 x 500	05896-11

COSMOSIL15SL-II Guard Column

Column Size I.D. x Length (mm)	Product Number
28 x 50	05892-51
50 x 50	05894-31

● Fast LC column (Particle Size: 3 µm)

COSMOSIL 3SL-II Packed Column

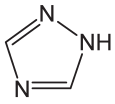
Column Size I.D. x Length (mm)	Product Number
4.6 x 10	38059-61
4.6 x 50	38060-21
4.6 x 100	38061-11

7. Hydrophilic Interaction Chromatography Column

COSMOSIL HILIC

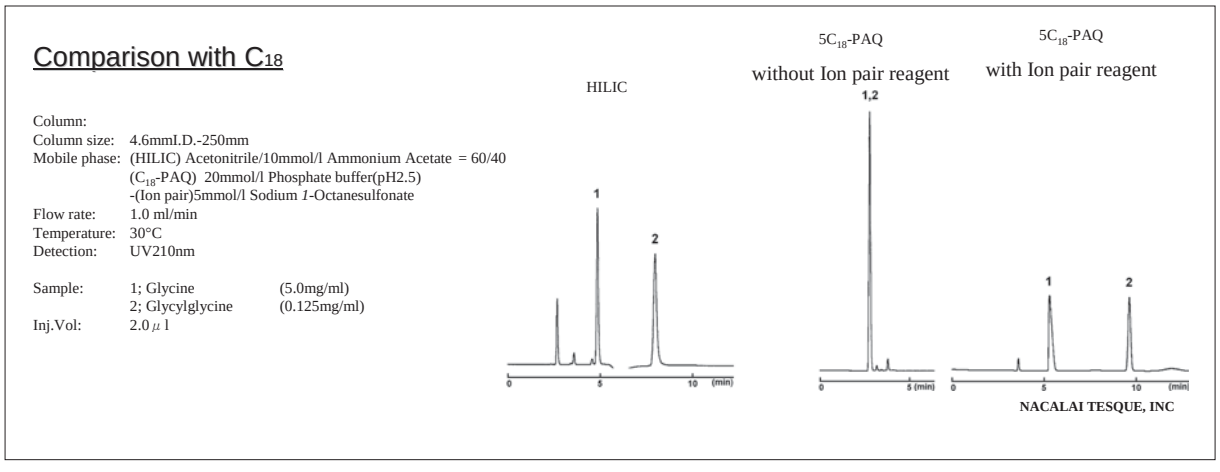
- Triazole bonded stationary phase
- Enhanced hydrophilic interaction
- Excellent retention for highly polar analytes
- Unique anion-exchange mechanism

Specifications

Packing Material	HILIC
Silica Gel	High Purity Porous Spherical Silica
Average Particle Size	5 µm
Average Pore Size	approx. 120 Å
Specific Surface Area	approx. 300 m^2/g
Bonded Phase Stracture	
Bonded phase	Triazole
Interaction	Hydrophilic Interaction, Anion Exchange
Object substance	Hydrophilic Compounds, Acidic Compounds
Features	Suitable for non-retaining by C_{18}

Comparison with C_{18}

The hydrophilic interaction chromatography is a variation of normal phase chromatography where a polar stationary phase is used with a mobile phase which contains a high concentration of water miscible organic solvent and a low concentration of aqueous eluent. The main retention mechanism is the partitioning of the polar analytes between the polar stationary and the non-polar mobile phase. As it is also called “aqueous normal phase”, the elution order is similar to that of normal phase and the sample elution is in the order of increasing hydrophilicity. Without using ion-pair reagent COSMOSIL HILIC retains highly polar analytes that would not be retained in reversed phase chromatography. It also shows a weak anion-exchange mechanism with the positively charged stationary phase, thus acidic compound is strongly retained.



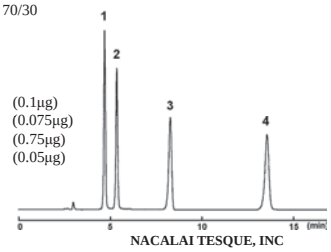
Applications

●Melamine Related Compunds

COSMOSIL Application Data

Column: HILIC
Column size: 4.6mmI.D.-250mm
Mobile phase: Acetonitrile/ 10mmol/l Ammonium Acetate = 70/30
Flow rate: 1.0 ml/min
Temperature: 30°C
Detection: UV225nm

Sample: 1; Melamine (0.1µg)
2; Ammeline (0.075µg)
3; Cyanuric Acid (0.75µg)
4; Ammelide (0.05µg)



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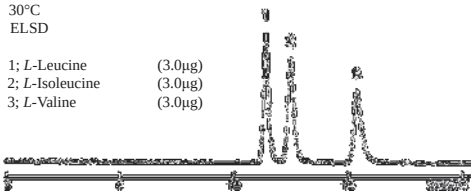
●BCAA (amino acid branhed-chain)

COSMOSIL Application Data

Column: HILIC
Column size: 4.6mmI.D.-250mm
Mobile phase: Acetonitrile/ 10mmol/l Ammonium Acetate = 85/15
Flow rate: 1.0 ml/min
Temperature: 30°C
Detection: ELSD

Sample: 1; *L*-Leucine (3.0µg)
2; *L*-Isoleucine (3.0µg)
3; *L*-Valine (3.0µg)



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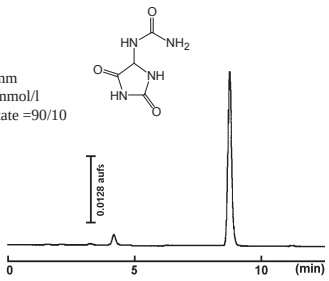
Optimizing Analytical Conditions

The COSMOSIL HILIC Chromatogram Index, which includes 167 chromatograms using COSMOSIL HILIC, is available on the NACALAI TESQUE website at http://www.nacalai.co.jp/en/cosmosil/. This index is a useful tool for all chromatographers, assisting them to optimize analytical conditions for hydrophilic interaction chromatography.

COSMOSIL Chromatogram Index

Sample: Allantoin
CAS No.: [97-59-6]
Molecular formula: $C_4H_6N_4O_3$
Column: HILIC
Column size: 4.6mmI.D.-250mm
Mobile phase: Acetonitrile/ 10mmol/l Ammonium acetate =90/10
Flow rate: 1.0 ml/min
Temperature: 30°C
Detection: UV220 nm
Attenuation: 0.128 aufs
Sample conc.: 1.0mg/ml
Injection volume: 1.0µl
Retention time: 8.75min
Capacity factor: 2.02



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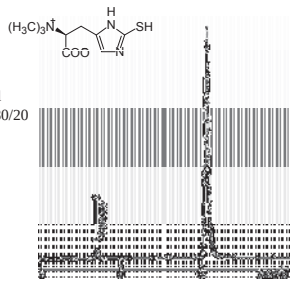
COSMOSIL Chromatogram Index

Sample: *L*-(+)-Ergothioneine
CAS No.: [497-30-3]
Molecular formula: $C_9H_{15}N_3O_2S$
Column: HILIC
Column size: 4.6mmI.D.-250mm
Mobile phase: Acetonitrile/ 10mmol/l Ammonium acetate =80/20
Flow rate: 1.0 ml/min
Temperature: 30°C
Detection: UV220 nm
Attenuation: 0.128 aufs
Sample conc.: 1.0mg/ml
Injection volume: 1.0µl
Retention time: 10.29min
Capacity factor: 2.79



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Ordering Information

● Analytical / Preparative Column (Particle Size: 5 µm)

COSMOSIL HILIC Packed Column

Column Size I.D. x Length (mm)	Product Number
1.0 x 150	07869-11
1.0 x 250	07870-71
2.0 x 30	08568-21
2.0 x 50	07052-91
2.0 x 100	08569-11
2.0 x 150	07054-71
2.0 x 250	07489-91
3.0 x 150	07871-61
3.0 x 250	07872-51

Column Size I.D. x Length (mm)	Product Number
4.6 x 150	07056-51
4.6 x 150 3 lots set*	09385-23
4.6 x 250	07057-41
10 x 250	07059-21
20 x 250	07060-81
28 x 250	07875-21

*For 4.6 x 150 3 lots set, please refer to page 11.

COSMOSIL HILIC Guard Column

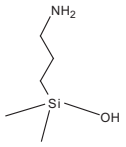
Column Size I.D. x Length (mm)	Product Number
4.6 x 10	07055-61
10 x 20	07058-31
20 x 20	07854-91
20 x 50	07873-41
28 x 50	07874-31

8. Mono- and Oligosaccharide Analysis Columns

Introduction

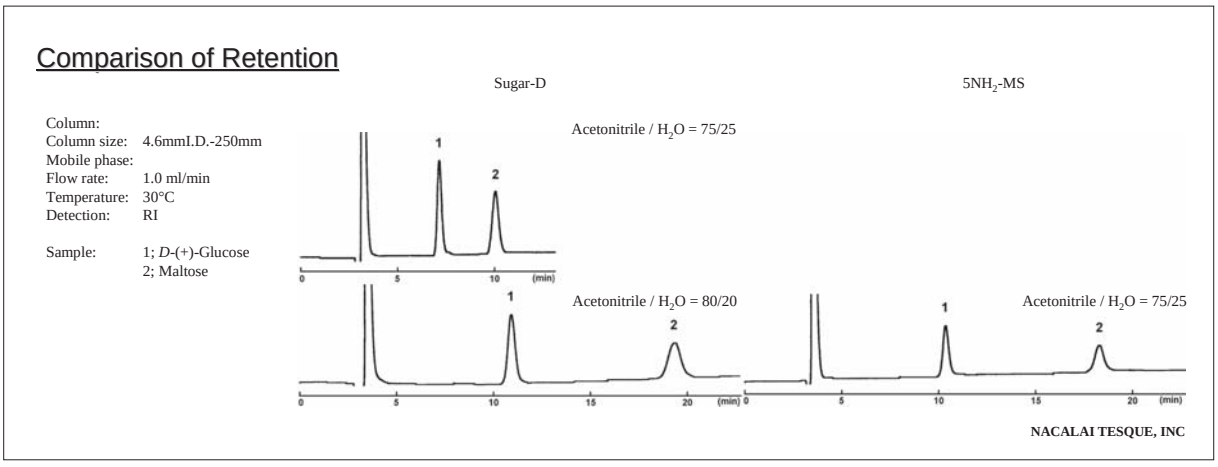
Saccharides are not retained on standard C_{18} columns because of the low hydrophobicity of compounds. COSMOSIL Sugar-D and NH_2-MS are specifically designed for separation of saccharides. COSMOSIL C_{18}-PAQ is recommended for hydrophobic glycosides or saccharide derivatives.

Specifications

Packing Material	Sugar-D	NH_2-MS
Silica Gel	High Purity Porous Spherical Silica	
Average Particle Size	5 µm	
Average Pore Size	—	approx. 120 Å
Specific Surface Area	—	approx. 300 m^2/g
Bonded Phase Stracture	—	
Bonded Phase	Secondary/Tertiary Amine	Aminopropyl Group
Bonding Type	—	Polymeric
Object Substances	Monosaccharides, Oligosaccharides	
End-capping Treatment	—	Near-perfect Treatment
Carbon Load	—	approx. 4%
Features	・First choise of saccharide analysis ・High durability ・Good quantitative analysis	・Different selectivity from Sugar-D

Comparison of Retention

The conventional aminopropyl column is slightly more retentive than Sugar-D. The retention time can be adjusted by increasing the concentration of acetonitrile in the mobile phase by 5%-10%

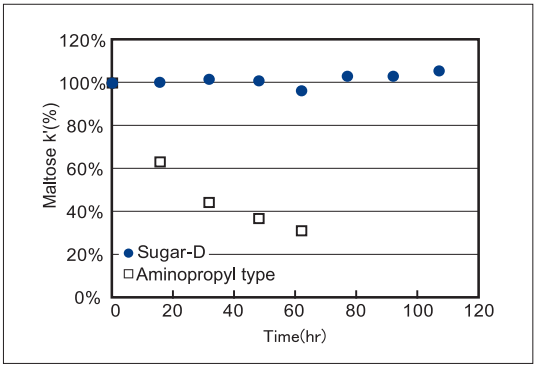


COSMOSIL Sugar-D

- Novel stationary phase for saccharides
- Superior durability to conventional amino columns
- Minimized undesirable adsorption

Comparison of Durability

The decrease of retention time was compared between COSMOSIL Sugar-D and conventional aminopropyl bonded stationary phase under severe condition of 100% water as eluent between tests. The capacity factor did not decrease in case of COSMOSIL Sugar-D.

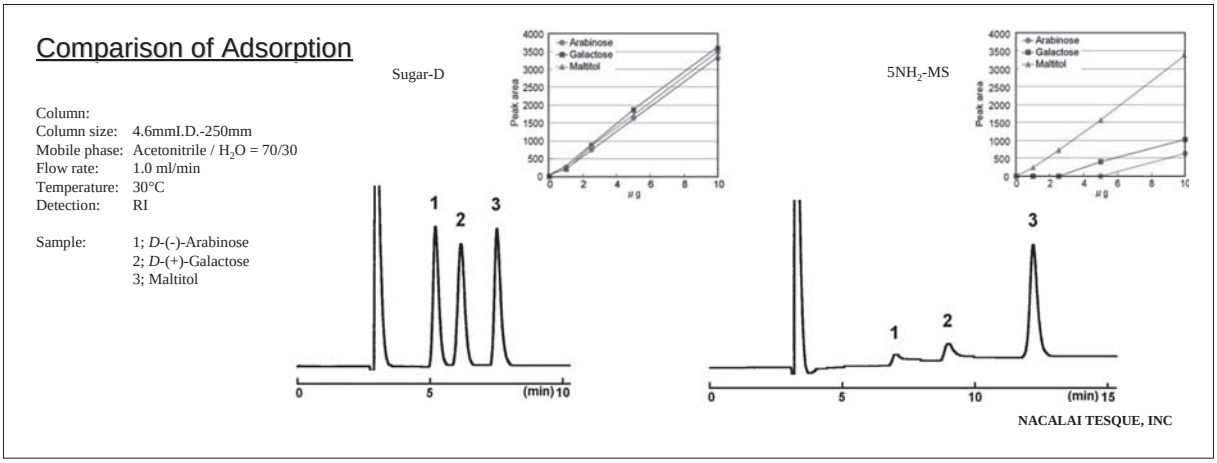


Decomposition Condition

Solution	Water
Flow Rate	1.0 ml/min
Temperature	Room Temperature
Column	4.6 mm I.D. x 250 mm
Mobile Phase	Acetonitrile : Water = 70 : 30
Flow Rate	1.0 ml/min
Temperature	30°C
Detection	RI
Sample	Maltose

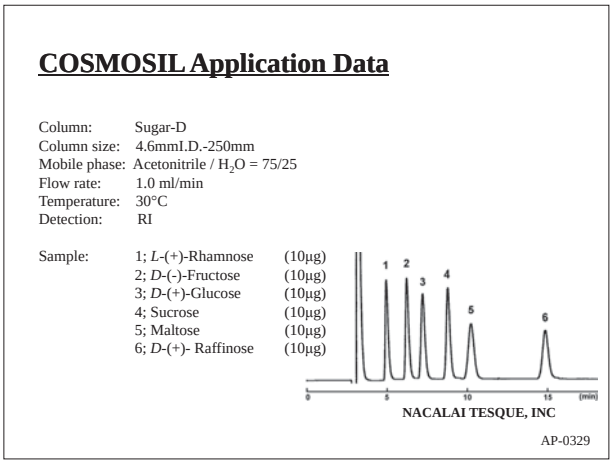
Comparison of Adsorption

Certain types of saccharides such as arabinose or galactose are partially or temporarily adsorbed on conventional aminopropyl stationary phases causing tailing or no elution at all. COSMOSIL Sugar-D provides superior separation and high recovery for these saccharides.

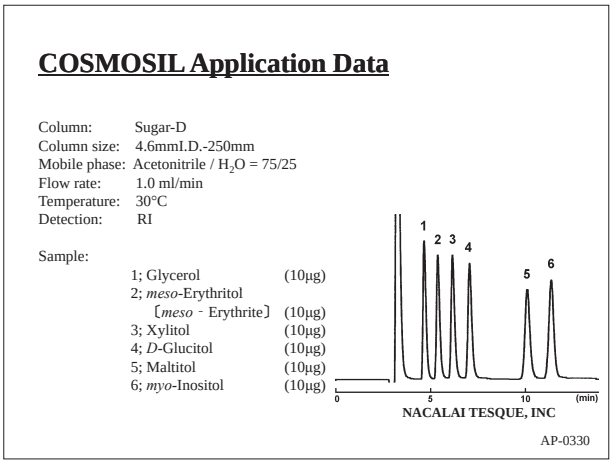


Applications

●Mono- and Oligosaccharides



●Polyols



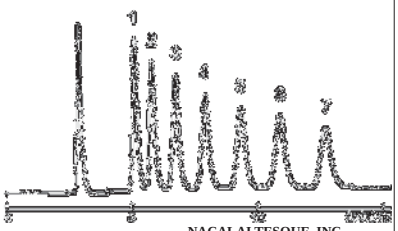
Applications

●Oligomaltoses

COSMOSIL Application Data

Column: Sugar-D
Column size: 4.6mmI.D.-250mm
Mobile phase: Acetonitrile / H_2O = 65/35
Flow rate: 1.0 ml/min
Temperature: 30°C
Detection: RI

Sample:
1; *D*-(+)-Glucose (10μg)
2; Maltose (10μg)
3; Maltotriose (10μg)
4; Maltotetraose (10μg)
5; Maltopentaose (10μg)
6; Maltohexaose (10μg)
7; Maltoheptaose (10μg)



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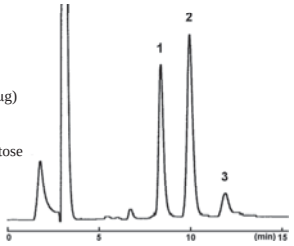
●Oligofructoses

COSMOSIL Application Data

Column: Sugar-D
Column size: 4.6mmI.D.-250mm
Mobile phase: Acetonitrile / H_2O = 70/30
Flow rate: 1.0 ml/min
Temperature: 30°C
Detection: RI

Sample: Fructooligosaccharides (50μg)
1; *1*-Kestose
2; Nystose
3; *1*-Fructofuranosyl-*D*-nystose



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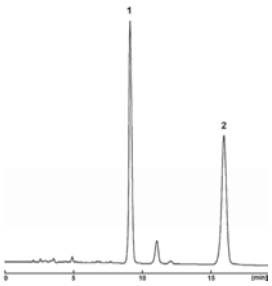
AP-0320

●Sweeteners

COSMOSIL Application Data

Column: Sugar-D
Column size: 4.6mmI.D.-250mm
Mobile phase: Acetonitrile/ H_2O = 85/15
Flow rate: 1.0 ml/min
Temperature: 30°C
Detection: UV210nm

Sample: 1; Stevioside
2; Rebaudioside A



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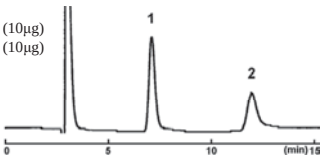
●Amino sugars

COSMOSIL Application Data

Column: Sugar-D
Column size: 4.6mmI.D.-250mm
Mobile phase: Acetonitrile/ 20mmol/l Phosphate buffer(pH7) = 70/30
Flow rate: 1.0 ml/min
Temperature: 30°C
Detection: RI

Sample: 1; *D*-(+)-Mannose (10μg)
2; *D*-Mannosamine (10μg)



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AP-0333

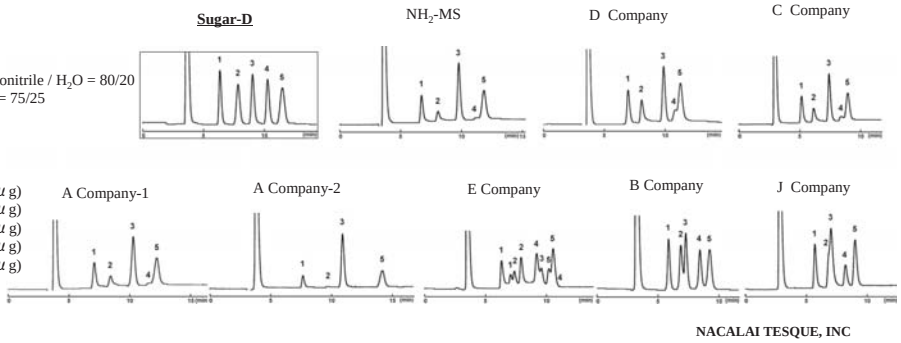
Comparison with Other Company Columns

The separation and the adsorption of monosaccharides were compared using COSMOSIL Sugar-D and other companies columns. Separation of aldoses, containing aldehyde group per molecule, is usually problematic with undesirable adsorption. COSMOSIL Sugar-D provides excellent separations for these saccharides.

Comparison

Column:
Column size: 4.6mmI.D.-250mm
Mobile phase: (Sugar-D, E Company) Acetonitrile / H_2O = 80/20
(Others) Acetonitrile / H_2O = 75/25
Flow rate: 1.0 ml/min
Temperature: 30°C
Detection: RI

Sample:
1; *L*-(+)-Rhamnose (10 μ g)
2; *D*-(+)-Xylose (10 μ g)
3; *D*-(-)-Fructose (10 μ g)
4; *D*-(+)-Mannose (10 μ g)
5; *D*-(+)-Glucose (10 μ g)



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Ordering Information

● Analytical / Preparative Column

COSMOSIL Sugar-D Packed Column

Column Size I.D. x Length (mm)	Product Number
2.0 x 250	05689-31
3.0 x 150	05690-91
3.0 x 250	05691-81

Column Size I.D. x Length (mm)	Product Number
4.6 x 150	05395-71
4.6 x 250	05397-51
10 x 250	05692-71
20 x 250	05693-61

COSMOSIL Sugar-D Guard Column

Column Size I.D. x Length (mm)	Product Number
4.6 x 10	05394-81
10 x 20	05696-31
20 x 50	05694-51

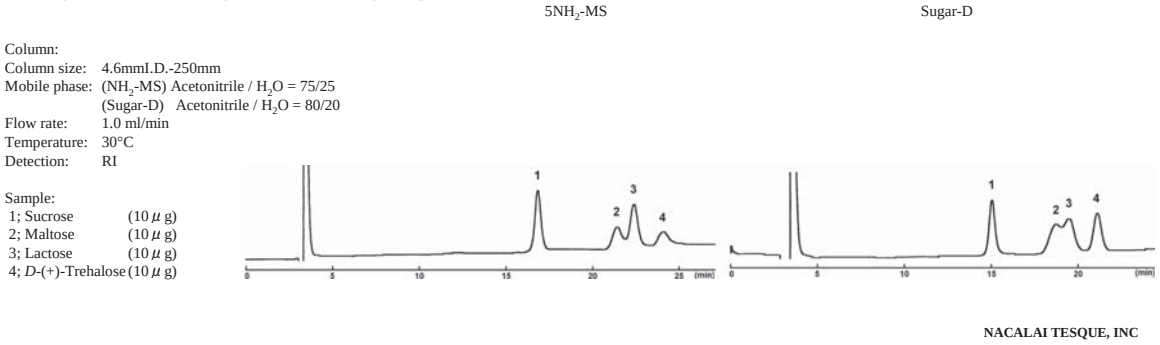
NH_2-MS

- Aminopropyl bonded stationary phase
- Different selectivity with Sugar-D

Comparison of Adsorption

NH_2-MS offer better separation than Sugar-D in some cases depending on samples

Comparison of Separation Property



Ordering Information

● Analytical / Preparative Column (Particle Size: 5 μm)

COSMOSIL 5NH_2-MS Packed Column

Column Size I.D. x Length (mm)	Product Number
4.6 x 150	38245-11
4.6 x 250	38246-01

Column Size I.D. x Length (mm)	Product Number
10 x 250	38249-71
20 x 250	38250-31

COSMOSIL 5NH_2-MS Guard Column

Column Size I.D. x Length (mm)	Product Number
4.6 x 10	38241-51
10 x 20	38242-41
20 x 50	06093-91

9. Protein Separation Columns

(1) Reversed Phase Chromatography

COSMOSIL Protein-R

- Excellent separation
- High recovery rate
- Outstanding stability at low pH

Specifications

Packing Material	Protein-R
Silica Gel	High Purity Porous Spherical Silica
Average Particle Size	5 µm
Average Pore Size	approx. 300 Å
Specific Surface Area	approx. 150 m^2/g
Bonded Phase	Octadecyl Group
Bonding Type	Polymeric
Main Interaction	Hydrophobic Interaction
End-capping Treatment	Near-perfect Treatment
Features	・High recovery rate ・Acid-resistant

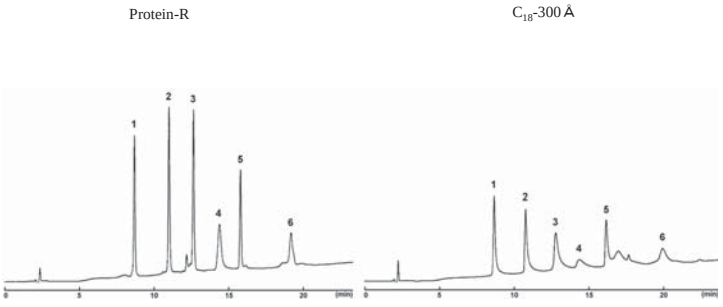
Comparison of Separation

Protein-R shows sharper peaks for proteins than conventional C_{18} wide pore columns.

Comparison of Separation Property

Column:
Column size: 4.6mmI.D.-150mm
Mobile phase: A; 0.05%TFA-20%Acetonitrile
B; 0.05%TFA-60%Acetonitrile
B conc. 0→100% 20min Linear gradient
Flow rate: 1.0 ml/min
Temperature: 30°C
Detection: UV220nm

Sample:
1; Ribonuclease A (1.0 µg)
2; Cytochrome C (1.0 µg)
3; Lysozyme (1.0 µg)
4; Albumin, Bovine [BSA] (1.0 µg)
5; Myoglobin (1.0 µg)
6; Albumin, Egg [Ovalbumin] (1.0 µg)



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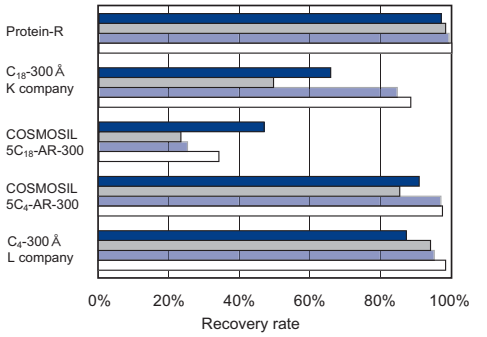
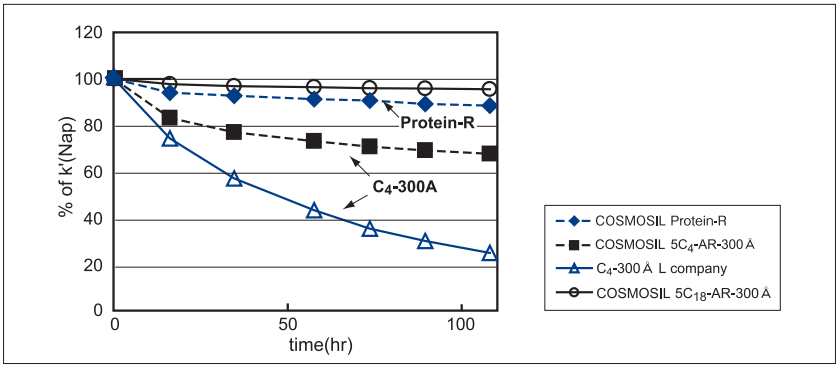


Recovery Rate

The figure left shows recovery rates for proteins using different columns. Protein-R shows a higher recovery rate than C_4-300 and a much higher recovery rate than C_{18}-300.

Comparison of Durability Against Acidic Mobile Phase



The figure left shows durability against acidic mobile phase of various columns. Protein-R shows a higher acid durability than C_4-300.

Degradation test with 0.1%-Trifluoroacetic Acid at 60℃
(k'): Napthalene in the mobile phase (Methanol : Water = 50 : 50)

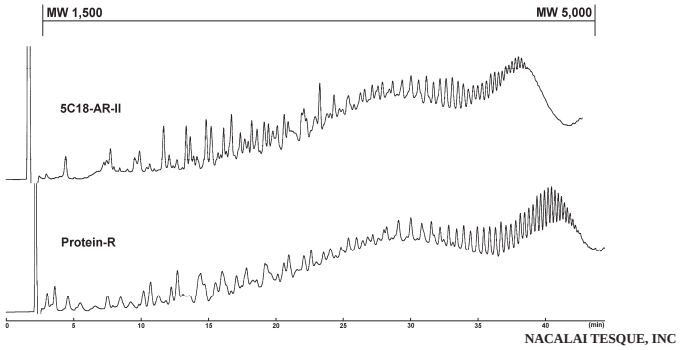
Application of Peptide Separation

$5C_{18}$-MS-II (pore size 120 Å) shows better separation of low-molecular weight proteins, but Protein-R shows better separation of high-molecular weight proteins.

COSMOSIL Application Data

Column:
Column size: 4.6mmI.D.-150mm
Mobile phase: A; 0.05%TFA-H_2O
B; 0.05%TFA-Acetonitrile
B conc. 0→35% 60min Linear gradient
Flow rate: 1.0 ml/min
Temperature: 30°C
Detection: UV220nm

Sample: Poly-L-Glutamic Acid Sodium Salt
(MW 1500-5000) (200µg)



Ordering Information

● Analytical / Preparative Column (Particle Size: 5 µm)

COSMOSIL Protein-R Packed Column

Column Size I.D. x Length (mm)	Product Number
2.0 x 150	06514-71
4.6 x 50	06525-31
4.6 x 150	06526-21
4.6 x 250	06527-11

Column Size I.D. x Length (mm)	Product Number
10 x 150	06529-91
10 x 250	06530-51
20 x 150	06531-41
20 x 250	06532-31

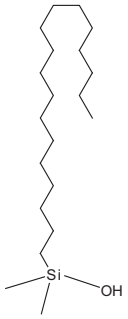
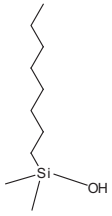
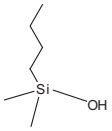
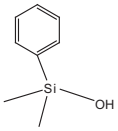
COSMOSIL Protein-R Guard Column

Column Size I.D. x Length (mm)	Product Number
4.6 x 10	06518-31
10 x 20	06528-01
20 x 20	08692-81

COSMOSIL C_{18}-AR-300, C_8-AR-300, C_4-AR-300, Ph-AR-300

- Wide pore reversed phase column
- 4 types of phases (octadecyl, octyl, butyl and phenyl)

Specifications

Packing Material	5C_{18}-AR-300	5C_8-AR-300	5C_4-AR-300	5Ph-AR-300
Silica Gel	High Purity Porous Spherical Silica			
Average Particle Size	5 µm			
Average Pore Size	approx. 300 Å			
Specific Surface Area	approx. 150 m^2/g			
Bonded Phase Stracture				
Bonded Phase	Octadecyl Group	Octyl Group	Butyl Group	Phenyl Group
Bonding Type	Polymeric			
Main Interaction	Hydrophobic Interaction			Hydrophobic Interaction π-π Interaction
End-capping Treatment	Near-perfect Treatment			
Carbon Load	approx. 12%	approx. 7%	approx. 6%	approx. 7%

Comparison of Separation

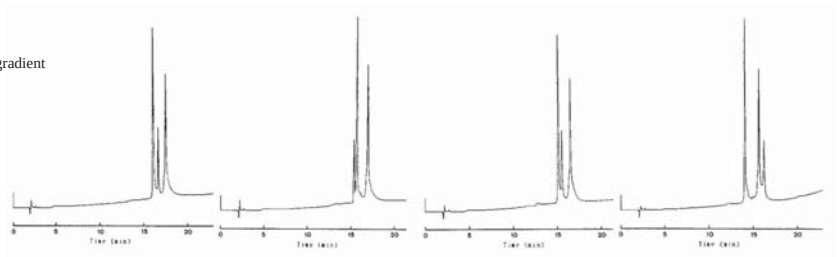
COSMOSIL AR-300 packed column series offer 3 types of alkyl phases and a phenyl phase.

Comparison of Separation Property

5C_{18}-AR-300 5C_8-AR-300 5C_4-AR-300 5Ph-AR-300

Column:
Column size: 4.6mmI.D.-150mm
Mobile phase: A; 0.05%TFA-20%Acetonitrile
B; 0.05%TFA-60%Acetonitrile
B conc. 0→100% 20min Linear gradient
Flow rate: 1.0 ml/min
Temperature: 30°C
Detection: UV220nm

Sample: Hemoglobin, Bovine (10 µg)



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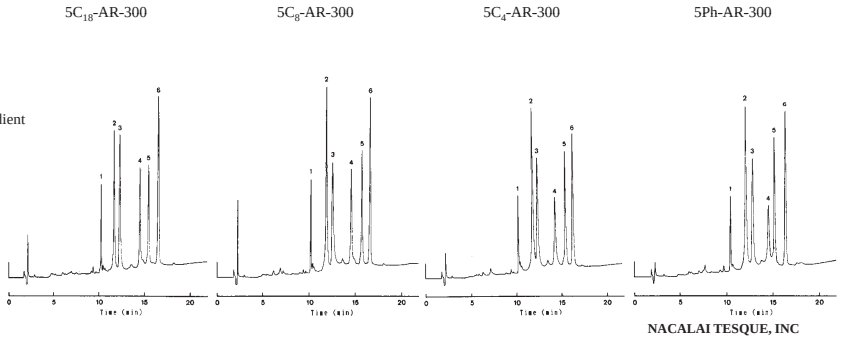
COSMOSIL Application Data

Column:
Column size: 4.6mmI.D.-150mm
Mobile phase: A; 0.05%TFA-20%Acetonitrile
B; 0.05%TFA-60%Acetonitrile
B conc. 0→100% 20min Linear gradient
Flow rate: 1.0 ml/min
Temperature: 30°C
Detection: UV220nm, 0.64AUFS

Sample:
1; Insulin (1.5μg)
2; Trypsinogen (6.0μg)
3; Transferrin (4.0μg)
4; Trypsin Inhibitor (5.0μg)
5; α-Chymotrypsinogen A (4.0μg)
6; Carbonic Anhydrase (3.0μg)



Ordering Information

● Analytical / Preparative Column (Particle Size: 5 μm)

COSMOSIL 5C18-AR-300 Packed Column

Column Size I.D. x Length (mm)	Product Number
4.6 x 50	37911-01
4.6 x 150	37913-81
4.6 x 250	37914-71

Column Size I.D. x Length (mm)	Product Number
10 x 150	37917-41
10 x 250	37918-31
20 x 150	37919-21
20 x 250	37920-81

COSMOSIL 5C18-AR-300 Guard Column

Column Size I.D. x Length (mm)	Product Number
4.6 x 10	37910-11
10 x 20	37965-11

COSMOSIL 5C8-AR-300 Packed Column

Column Size I.D. x Length (mm)	Product Number
4.6 x 50	37951-81
4.6 x 150	37953-61
4.6 x 250	37954-51

Column Size I.D. x Length (mm)	Product Number
10 x 150	34345-21
10 x 250	34247-11
20 x 150	05861-51
20 x 250	34364-71

COSMOSIL 5C8-AR-300 Guard Column

Column Size I.D. x Length (mm)	Product Number
4.6 x 10	37950-91
10 x 20	34464-61

COSMOSIL 5C4-AR-300 Packed Column

Column Size I.D. x Length (mm)	Product Number
4.6 x 50	37956-31
4.6 x 150	37958-11
4.6 x 250	37959-01

Column Size I.D. x Length (mm)	Product Number
10 x 150	34249-91
10 x 250	38047-11
20 x 150	34477-01
20 x 250	38048-01

COSMOSIL 5C4-AR-300 Guard Column

Column Size I.D. x Length (mm)	Product Number
4.6 x 10	37955-41
10 x 20	05862-41

COSMOSIL 5Ph-AR-300 Packed Column

Column Size I.D. x Length (mm)	Product Number
4.6 x 50	37961-51
4.6 x 150	37963-31
4.6 x 250	37964-21

Column Size I.D. x Length (mm)	Product Number
10 x 150	05865-11
10 x 250	34267-51
20 x 150	05866-01
20 x 250	34468-21

COSMOSIL 5Ph-AR-300 Guard Column

Column Size I.D. x Length (mm)	Product Number
4.6 x 10	37960-61
10 x 20	34268-41

(2) Gel Filtration Chromatography Column (aqueous)

COSMOSIL Diol-120-II,Diol-300-II

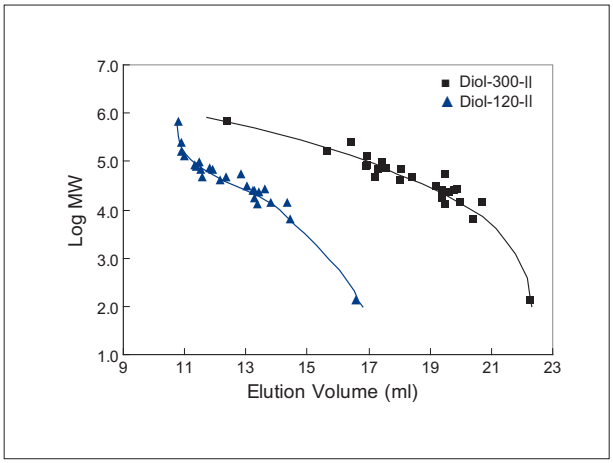
- Ideal for the size-based separation of proteins and water soluble polymers
- Reduce undesirable adsorption

Specifications

Packing Material	5Diol-120-II	5Diol-300-II
Silica Gel	High Purity Porous Spherical Silica	
Average Particle Size	5 µm	
Average Pore Size	approx. 120 Å	approx. 300 Å
Bonded Phase	Diol Group	
Object Substance	Proteins, Water Soluble Polymers	
Flow Rate	0.5-1.0 (ml/min)	
Selection of Pore Size (protein)	5,000-100,000	10,000-700,000
Selection of Pore Size (water soluble polymers)	300-30,000	500-300,000

Calibration Curve

- Calibration Curve of Proteins



Column COSMOSIL 5Diol-II 7.5mmI.D.x 600 mm
Mobile Phase 20mmol/l Phosphate Buffer (pH7.0)+100mmol/l Na_2S0_4
Flow Rate 1.0ml/min
Temperature 30°C

Sample	M.W.	Sample	M.W.
Thyroglobulin	660,000	Peroxidase	40,000
Catalase	250,000	Carbonic Anhydrase	30,000
Glucose Oxidase	160,000	α-Chymotrypsinogen A	25,700
Uricase	128,000	α-Chymotrypsin	25,200
Choline Oxidase	95,000	Trypsinogen	24,000
Transferrin	85,000	Trypsin (bovine)	23,300
Conalbumin	77,500	Myoglobin	17,000
Malate Dehydrogenase	70,000	Lysozyme	14,300
α-Glucosidase	68,500	Ribonuclease A	13,700
Albumin (BSA)	66,000	Cytochrome C	12,400
α-Amylase	52,500	Aprotinin	6,500
Fetuin	48,000	Gly-Gly	132
Albumin (Ovalbumin)	45,000		

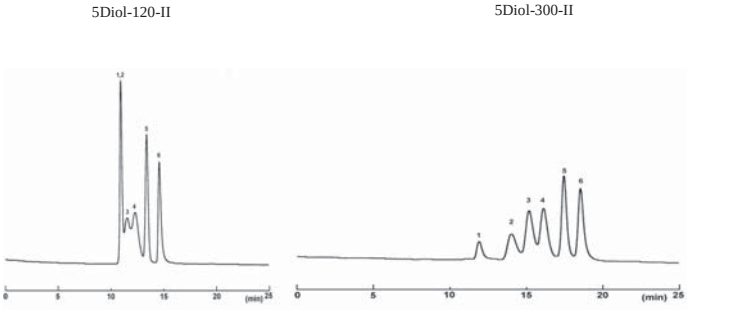
Separation of Standard Proteins

COSMOSIL Diol-II series are available in two different pore sizes, Diol-120-II and Diol-300-II. Combination of these two columns enables separation of a wide M.W. range of samples.

Comparison of Separation Property

Column:
Column size: 7.5mmI.D.-600mm
Mobile phase: 20mmol/l Phosphate buffer(pH 7.0)
+100mmol/l Na_2SO_4
Flow rate: 1.0 ml/min
Temperature: Room temperature
Detection: UV220nm

Sample: 1; Thyroglobulin
2; Glucose Oxidase
3; Conalbumin
4; Peroxidase
5; Myoglobin
6; Aprotinin



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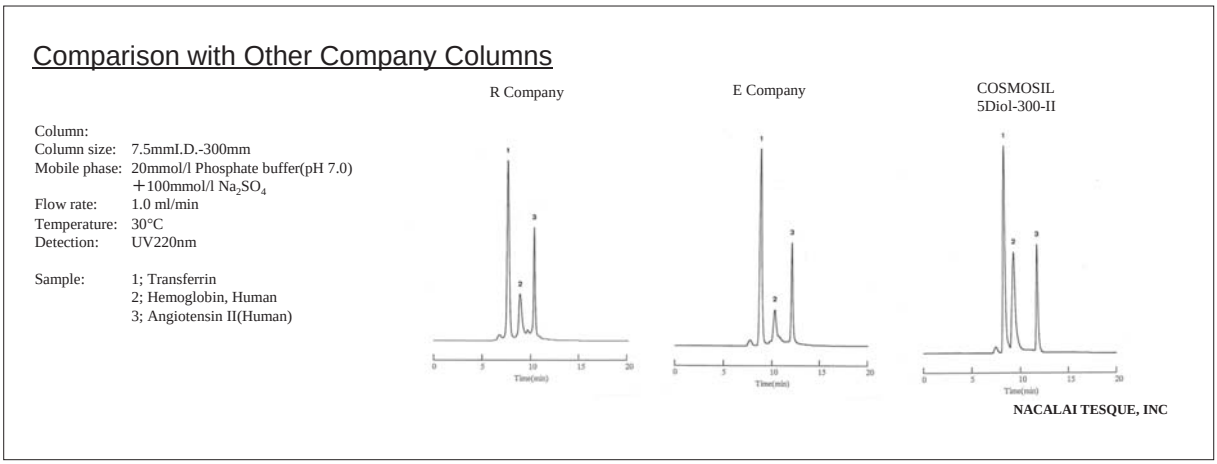
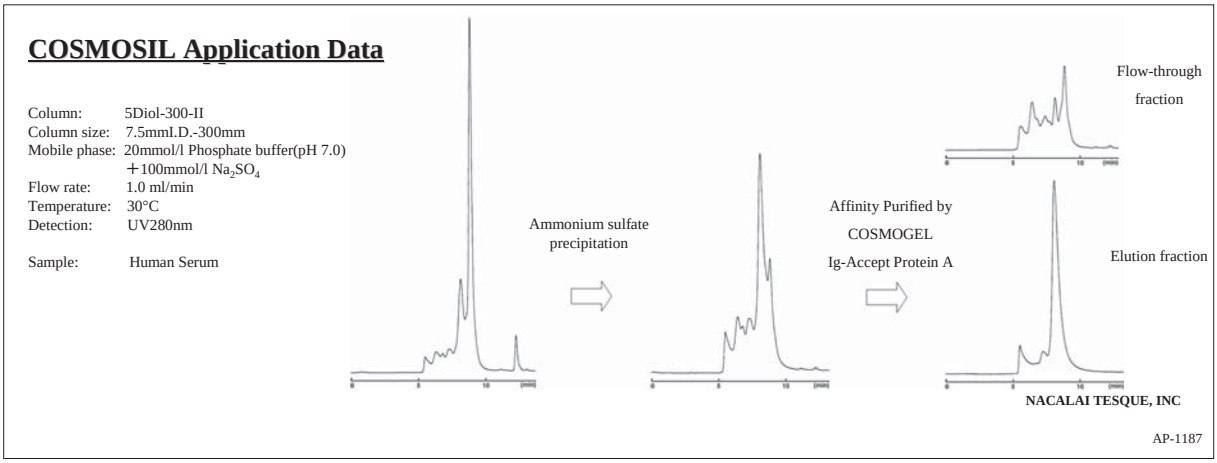
Comparison with Other Company Columns



Applications

●Human Serum



Ordering Information

● Analytical / Preparative Column (Particle Size: 5 µm)

COSMOSIL 5Diol-120-II Packed Column

Column Size I.D. x Length (mm)	Product Number
7.5 x 300	38050-51
7.5 x 600	38051-41

COSMOSIL 5Diol-120-II Guard Column

Column Size I.D. x Length (mm)	Product Number
7.5 x 50	38049-91

COSMOSIL 5Diol-300-II Packed Column

Column Size I.D. x Length (mm)	Product Number
7.5 x 300	38053-21
7.5 x 600	38054-11

COSMOSIL 5Diol-300-II Guard Column

Column Size I.D. x Length (mm)	Product Number
7.5 x 50	38052-31

(3) Ion Exchange Chromatography Column

COSMOGEL IEX Series

- Available in 3 different ion-exchange modes
  (Anion-exchange type, Cation-exchange type, Amphoteric ion-exchange type)
- Available for 3 different application areas
  (for Purification, for Ultra-fast analysis, for Precise analysis)
- For separation of biopolymers such as proteins or nucleic acids

Specifications

Packing Material	Type Q	Type Q-N	Type S	Type S-N	Type M	Type M-N
Gel/Average Particle Size	Totally Porous Spherical Hydrophilic Polymer / 5 µm					
Average Pore Size	1000 Å	Non-porous	1000 Å	Non-porous	1000 Å	Non-porous
Functional Group	$-CH_3N^+(CH_3)_3$		$-(CH_2)_3SO_3^-$		$-CH_3N+(CH_3)_3$ + $-(CH_2)_3SO_3^-$	
Protein Binding Capacity	110-150 mg BSA/ml-resin	12-20 mg BSA/ml-resin	70-100 mg human IgG/ml-resin	10-18 mg human IgG/ml-resin	55-75 mg(BSA)/ml 35-50 mg(IgG)/ml	6-10 mg(BSA)/ml 5-9 mg(IgG)/ml
Column Size I.D. x Lenght (mm)	4.6-50	4.6-30 / 4.6-100	4.6-50	4.6-30 / 4.6-100	4.6-50	4.6-100
Column Material	PEEK					
Connection	Waters Type					

Type of Packing Material

COSMOGEL IEX Series are available in amphoteric ion-exchange type in which two kinds of packing materials are mixed, as well as in widely used anion-exchange type and cation-exchange type.

Type of Packing Material	Target Sample	Average Pore Size	
		Porous (1000 Å)	Non-porous
Anion-exchange Type	Acidic Proteins / DNA	Type Q	Type Q-N
Cation-exchange Type	Basic Proteins	Type S	Type S-N
Amphoteric ion-exchange Type	All Proteins	Type M	Type M-N

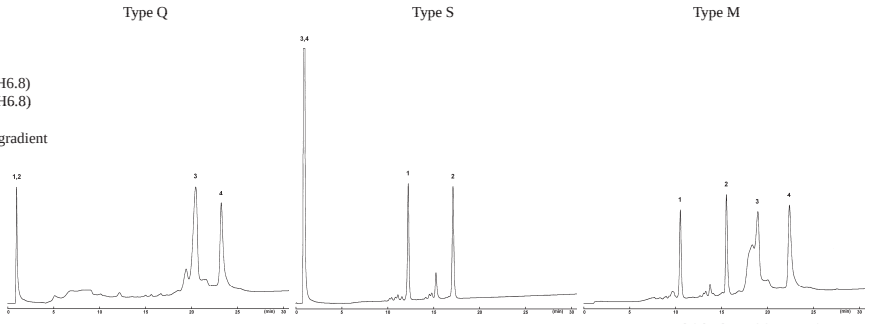
● Comprehensive isolation of proteins by Amphoteric ion-exchange type (Type M)

The amphoteric ion-exchange type enables the simultaneous separation of both acidic and basic proteins in one application.

COSMOSIL Application Data

Column: COSMOGEL IEX
Column size: 4.6mmI.D.-50mm
Mobile phase: A: 20mmol/l Phosphate buffer(pH6.8)
B: 20mmol/l Phosphate buffer(pH6.8)
+ 0.5mol/l NaCl
B conc. 0→100% 30min Linear gradient
Flow rate: 0.5 ml/min
Temperature: 30°C
Detection: UV220nm

Sample: 1; Ribonuclease A (1.5µg)
2; Cytochrome C (1.5µg)
3; Trypsin Inhibitor (6.0µg)
4; β-Galactosidase (6.0µg)



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Type of Column

COSMOGEL IEX Series are available for 3 types of applications:

Application	Pore Size	Column Size I.D. x Lenght (mm)	Column		
For Purification	Porous (1000 Å)	4.6-50	Type Q	Type S	Type M
For Precise Analysis	Non-porous	4.6-100	Type Q-N	Type S-N	Type M-N
For Ultra-fast Analysis	Non-porous	4.6-30	Type Q-N	Type S-N	—

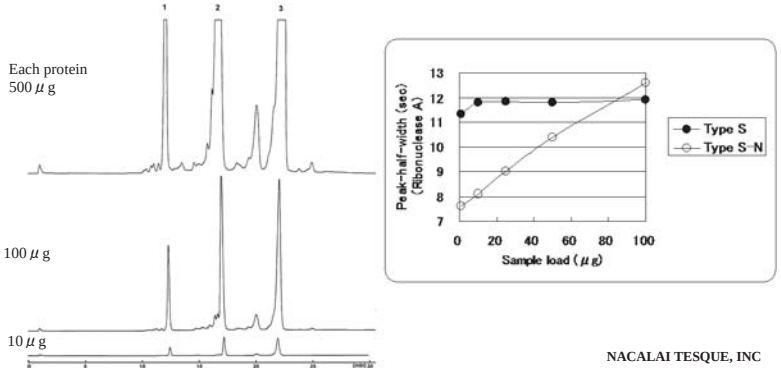
● For Purification: Type Q, Type S, Type M

The type of porous packing materials have higher binding capacity of proteins than the respective non-porous type, which means that peak shape does not spread even with injection of a large volume of sample. Therefore they are highly suitable for purification of large sample.

COSMOSIL Application Data

Column: COSMOGEL IEX Type S
Column size: 4.6mmI.D.-50mm
Mobile phase: A: 20mmol/l Phosphate buffer(pH6.8)
B: 20mmol/l Phosphate buffer(pH6.8) + 0.5mol/l NaCl
B conc. 0→100% 30min Linear gradient
Flow rate: 0.5 ml/min
Temperature: 30°C
Detection: UV280nm

Sample: 1; Ribonuclease A
2; Cytochrome C
3; Lysozyme



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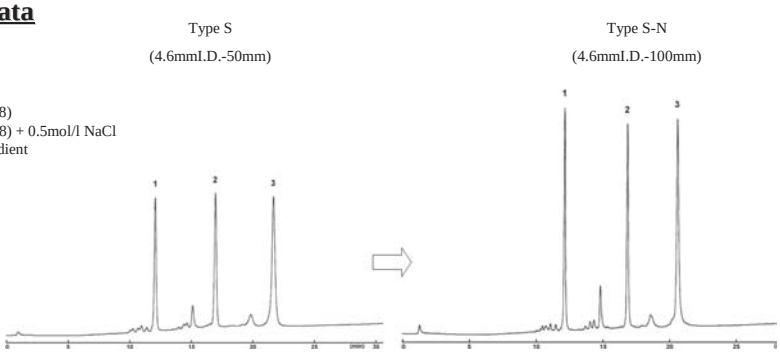
● For Precise Analysis: Type Q-N, Type S-N, Type M-N

The type of non-porous packing materials reduce spreading of samples in packing materials, result in high resolution separation for precise analysis such as quality control of antibody drugs. The longer column length also contributes to the sharper peaks.

COSMOSIL Application Data

Column: COSMOGEL IEX
Column size:
Mobile phase: A: 20mmol/l Phosphate buffer(pH6.8)
B: 20mmol/l Phosphate buffer(pH6.8) + 0.5mol/l NaCl
B conc. 0→100% 30min Linear gradient
Flow rate: 0.5 ml/min
Temperature: 30°C
Detection: UV220nm

Sample: 1; Ribonuclease A (1.65 μg)
2; Cytochrome C (1.65 μg)
3; Lysozyme (1.65 μg)



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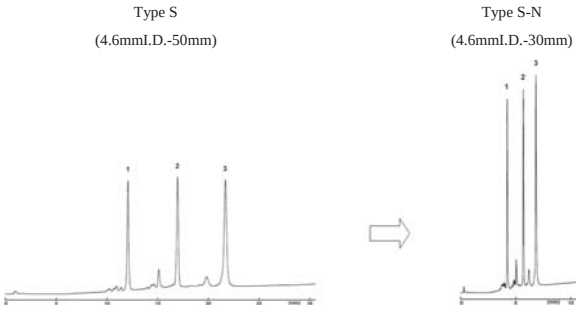
● For Ultra-fast Analysis: Type Q-N, Type S-N

The type of non-porous packing materials is not much affected by high flow rate and thus the materials are suitable for fast analysis. The shorter column length contributes to the fast analysis.

COSMOSIL Application Data

Column: COSMOGEL IEX
Column size:
Mobile phase: A: 20mmol/l Phosphate buffer(pH6.8)
B: 20mmol/l Phosphate buffer(pH6.8) + 0.5mol/l NaCl
B conc. 0→100% Linear gradient
Type S: 30min Type S-N: 10min
Flow rate: Type S: 0.5 ml/min Type S-N: 1.0 ml/min
Temperature: 30°C
Detection: UV220nm

Sample: 1; Ribonuclease A (1.65 μg)
2; Cytochrome C (1.65 μg)
3; Lysozyme (1.65 μg)



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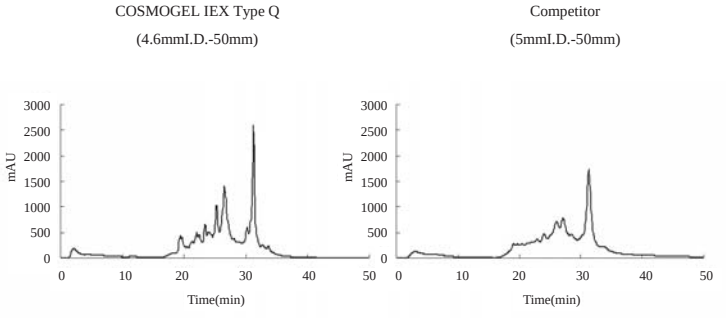
Applications

● Performance Comparison

COSMOSIL Application Data

Column:
Column size:
Mobile phase: A: 20mmol/l Tris buffer(pH8.3)
B: 20mmol/l Tris buffer(pH8.3) + 0.5mol/l NaCl
B conc. 0% (0-13min)
0→100% (13-43min)
100% (43-53min)
Flow rate: 0.5 ml/min
Temperature: 8°C
Detection: UV220nm
Equipment ÄKTAexplorer 10 S

Sample: Trypsin Inhibitor from Soybean (1mg/ml)
Inj.Vol. 500 μl



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Data courtesy of Kei WADA, Ph. D., Department of Biological Sciences, Graduate School of Science, Osaka University

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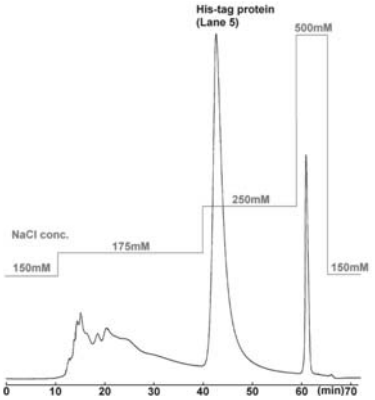
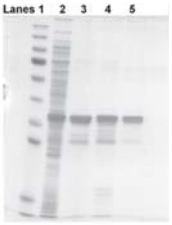
● Purification of His-tag Fusion Proteins

COSMOSIL Application Data

Column: COSMOGEL IEX Type Q
Column size: 4.6mmI.D.-50mm
Mobile phase: A: 20mmol/l Tris buffer(pH8.0)
B: 20mmol/l Tris buffer(pH8.0) + 0.5mol/l NaCl
B conc. 30% (0min)
35% (0-30min)
50% (30-50min)
100% (50-55min)
30% (55min-)
Flow rate: 0.5 ml/min
Temperature: room temperature
Detection: UV280nm

Sample: His-tag Protein in 20mmol/l Tris buffer(pH8.0) **(Lane 4)**
Inj.Vol. 5.0ml





Lane 1: Protein Makers for SDS-PAGE (# 09547-16)
Lane 2: His-tag protein [pET-41b(+)] in *E. Coli* lysate
Lane 3: Purified fraction by COSMOGEL His-Accept SF
Lane 4: Desalted fraction
Lane 5: Purified fraction by COSMOGEL IEX Type Q

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Ordering Information

Ion Exchange Mode	Product Name	Application	Column Size I.D. x Lenght (mm)	Product Number
Anion-exchange Type	COSMOGEL IEX Type Q	For Purification	4.6 x 50	06266-31
	COSMOGEL IEX Type Q-N	For Ultra-fast Analysis	4.6 x 30	06264-51
	COSMOGEL IEX Type Q-N	For Precise Analysis	4.6 x 100	06258-41
Cation-exchange Type	COSMOGEL IEX Type S	For Purification	4.6 x 50	06252-01
	COSMOGEL IEX Type S-N	For Ultra-fast Analysis	4.6 x 30	06251-11
	COSMOGEL IEX Type S-N	For Precise Analysis	4.6 x 100	06250-21
Amphoteric Ion-exchange Type	COSMOGEL IEX Type M	For Purification	4.6 x 50	06248-71
	COSMOGEL IEX Type M-N	For Precise Analysis	4.6 x 100	06244-11

COSMOSIL HIC

- Separate based on differences in hydrophobicity
- Little loss in enzyme activity and the tertiary structure of proteins

Specifications

Packing Material	HIC
Silica Gel	High Purity Porous Spherical Silica
Average Particle Size	5 µm
Average Pore Size	approx. 300 Å
Specific Surface Area	approx. 150 m^2/g
Main Interaction	Hydrophobic Interaction

Applications

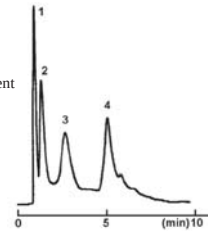
A buffer with high salt concentration, usually 1–2 mol/l of $(NH_4)_2SO_4$, is used as an initial mobile phase for adsorption of samples to a weakly hydrophobic stationary phase. The elution is done with a decreasing salt gradient. The application in lower left shows myoglobin elutes first than BSA under the buffer with high salt concentration, suggesting that myoglobin is less hydrophobic than BSA.

- Separation of Protein Standards

COSMOSIL Application Data

Column: HIC
Column size: 4.6mmI.D.-50mm
Mobile phase: A:20mmol/l Phosphate Buffer
+100mmol/l Na_2SO_4
+1.5mol/l $(NH_4)_2SO_4$(pH6.7)
B:20mmol/l Phosphate Buffer
+100mmol/l Na_2SO_4(pH6.7)
B conc. 0→100% 10min Linear gradient
Flow rate: 1.0 ml/min
Temperature: 30°C
Detection: UV220nm

Sample:
1; Myoglobin (1.0 µg)
2; β-Lactoglobulin (2.0 µg)
3; Hemoglobin, Bovine (5.0 µg)
4; Albumin, Bovine [BSA] (2.0 µg)



NACALAI TESQUE, INC

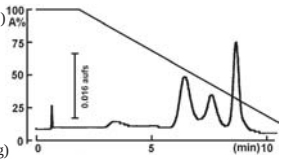
AP-0406

- Separation of β-Glucosidase

COSMOSIL Application Data

Column: HIC
Column size: 4.6mmI.D.-50mm
Mobile phase: A:20mmol/l Phosphate Buffer
+100mmol/l Na_2SO_4
+2mol/l $(NH_4)_2SO_4$(pH6.7)
B:20mmol/l Phosphate Buffer
+100mmol/l Na_2SO_4(pH6.7)
B conc. 0→100% 10min
Linear gradient
Flow rate: 1.0 ml/min
Temperature: 30°C
Detection: UV220nm

Sample: β-Glucosidase (15 µg)



NACALAI TESQUE, INC

AP-0407

Ordering Information

- Analytical / Preparative Column (Particle Size: 5 µm)

COSMOSIL 5HIC Packed Column

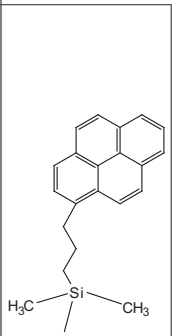
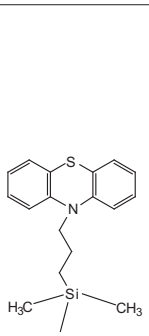
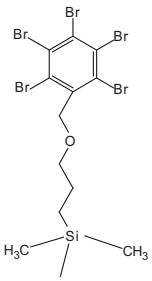
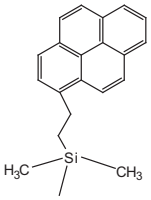
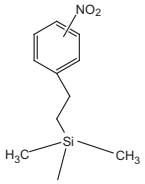
Column Size I.D. x Length (mm)	Product Number
4.6 x 50	04263-21

10. Columns for Fullerene Separation

Introduction

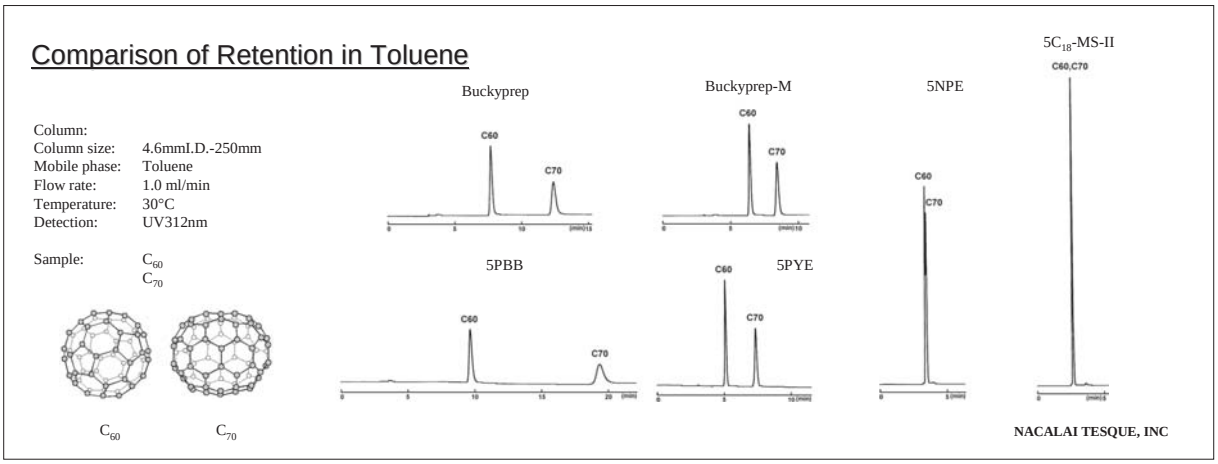
Separation of fullerenes, especially preparative scale separation, on conventional HPLC columns are always problematic due to the low solubility and low recovery rate of fullerenes. COSMOSIL offers a variety of columns designed for preparative scale separation of fullerenes including higher fullerenes, metallofullerenes and fullerene derivatives.

Specifications

Packing Material	Buckyprep	Buckyprep-M	PBB	PYE	NPE
Silica Gel	High Purity Porous Spherical Silica				
Average Particle Size	5 µm				
Average Pore Size	approx. 120 Å				
Specific Surface Area	approx. 300 m^2/g				
Bonded Phase Stracture					
Bonded Phase	Pyrenylpropyl Group	Phenothiazinyl Group	Pentabromobenzyl Group	Pyrenylethyl Group	Nitrophenylethyl Group
Bonding Type	Monomeric				
End-capping Treatment	Near-perfect Treatment	None	Near-perfect Treatment		
Carbon Load	approx. 17%	approx. 13%	approx. 8%	approx. 18%	approx. 9%
Features	• Standard column for fullerene separation.	• Designed to separate metallofullerenes	• Designded for preparative separation of C_{60}, C_{70}	• Separation of fullerene and structural isomers	• Separation of fullerene derivatives

Comparison of Retention

The figure below shows the retention time of C_{60} and C_{70} in toluene. Buckyprep, Buckyprep-M and PBB, and PYE nicely separate C_{60} and C_{70}. On the other hand, C_{18} with alkyl group and NPE can not separate them in toluene.



Solubility and Boiling Point of Each Solvent for C_{60}

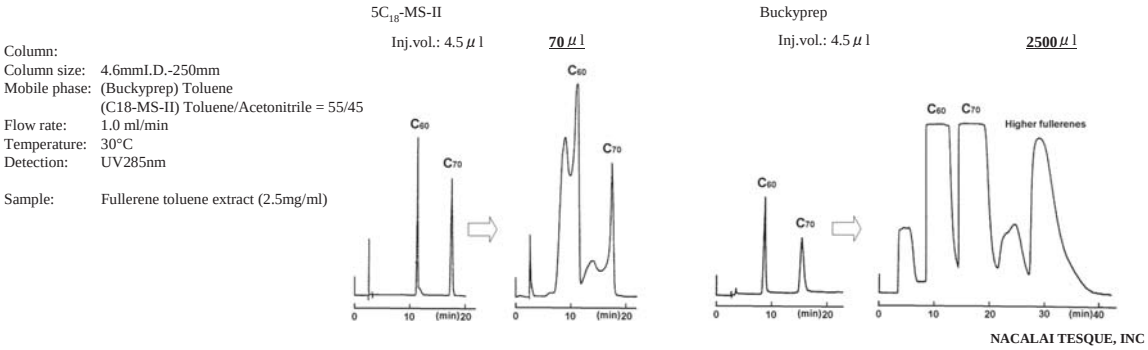
Solubility and boiling point of each solvent for C_{60}

Solvent	mg/ml	b.p. (°C)
Methanol	0.001	64.5
Acetonitrile	0.018	81.8
n-Hexane	0.046	68.7
Toluene	3.2	111
Chlorobenzene*	7.0	132
Carbon disulfide	12	46.3
1,2,4-Trichlorobenzene	21.3	213
o-Dichlorobenzene*	27	180

*R.S.Ruoff, et al., *J. phy. Chem.*, 97, 3379 (1993)

The table on the left shows solubility and boiling point of each solvent for C_{60}. Toluene is the most suitable solvent for due to its high solubility, appropriate boiling point, low cost and low corrosivity. Although C_{18} column with alkyl group can not separate C_{60} and C_{70} in toluene, by mixing acetonitrile and toluene, they can be separated as shown in the figure below. However, increase injection volume in preparative purification triggers tailing that result in practical injection volume limit of only 70 µl (175 µg). On the other hand, Buckyprep can separate well in toluene with non-tailing with the maximum injection volume of 2,500 µl (6.25 mg).

Difference in Preparative Separation



Suggested Solvents

● Features of suggested solvents for fellerene separation

Solvent	Feature
Chlorobenzene	Stronger the elution effect than toluene. Recommended for higher fullerenes.
o-Dichlorobenzene	Stronger the elution effect than chlorobenzene. Excellent solubility for fullerenes.
1,2,4-Trichlorobenzene	The strongest elution effect. It is sutable not only for mobile phase but also for washing solution for adsorbed higher fullerenes. In case of injecting continuously for high purity, recommend to wash the colums each purification . (4.6 mm I.D. x 250 mm) injection volume for washing: approx. 3ml (20 mm I.D. x 250 mm) injection volume for washing: approx. 50ml
Hexane	Weak elution effect. Recommende for weakly retained fullerenes.
Acetonitrile	Weak elution effect. Recommende for weakly retained fullerenes.

Note: Use them after filtration or distillation, if they are not for HPLC.

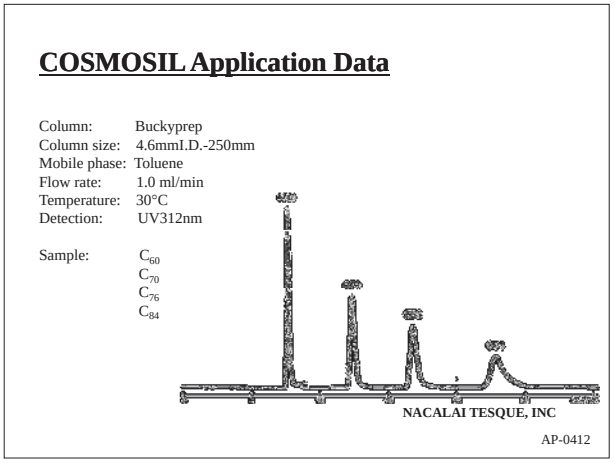
Except for alkali aqueous and strong acid solutions, other solvents can be used (water-free pyridine and others). Depending on solvents, pay attention to high pressure caused by high solvent viscosity.

COSMOSIL Buckyprep

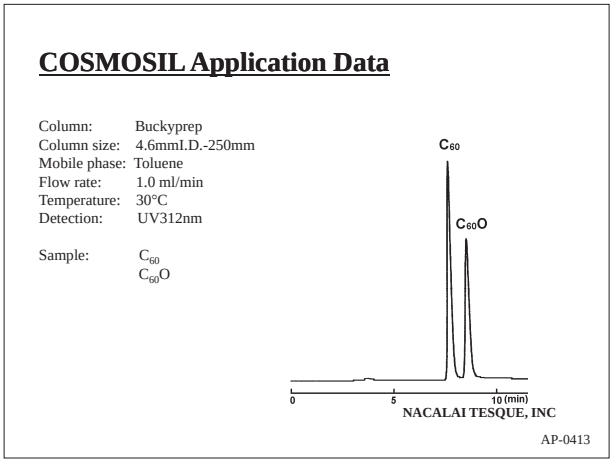
- Standard column for fullerene separation
- Excellent separation for higher and derivatized fullerenes

Applications

●Higher Fullerenes

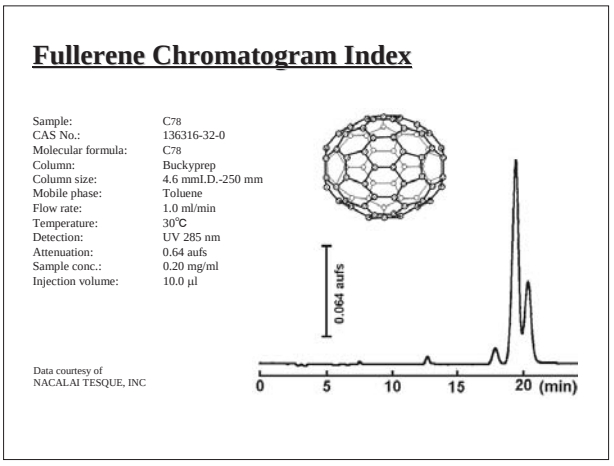


●Detivatized Fullerenes



Fullerene Chromatogram Index

Fullerene Chromatogram Index includes more than 100 chromatograms. If you are interested in this index, please feel free to e-mail us at info.intl@nacalai.com. The online version is available at the website of The Fullerenes, Nanotubes and Graphene Research Society below.



The Fullerenes, Nanotubes and Graphene Research Society
Website: http://fullerene-jp.org/jp/chromato_index_3.pdf

Ordering Information

● Analytical / Preparative Column (Particle Size: 5 µm)

COSMOSIL Buckyprep Packed Column

Column Size I.D. x Length (mm)	Product Number
4.6 x 250	37977-61
10 x 250	37981-91
20 x 250	37982-81
28 x 250	34346-11

COSMOSIL Buckyprep Guard Column

Column Size I.D. x Length (mm)	Product Number
4.6 x 10	37983-71
10 x 20	37984-61
20 x 50	34374-41
28 x 50	05871-21

COSMOSIL Buckyprep-M

- Different selectivity with Buckyprep
- Excellent separation for metallofullerenes

Applications

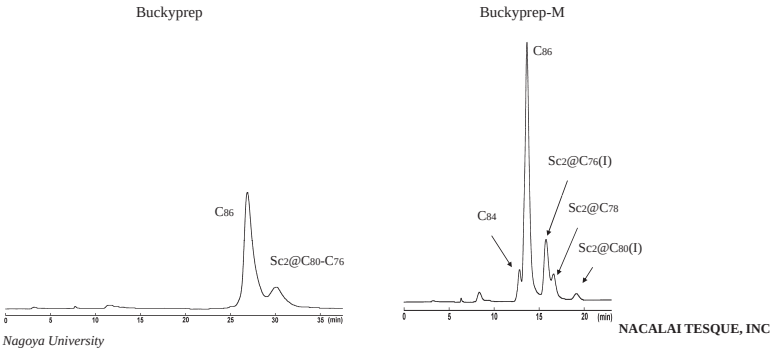
- Metallofullerenes

COSMOSIL Buckyprep-M is a phenothiazinyl group bonded silica based column specifically designed for metallofullerene separation. Metallofullerenes are retained more strongly than other fullerenes on this column. COSMOSIL Buckyprep-M is also effective for the separation of higher fullerenes and fullerene derivatives.

COSMOSIL Application Data

Column:
Column size: 4.6mmI.D.-250mm
Mobile phase: Toluene
Flow rate: 1.0 ml/min
Temperature: 30°C
Detection: UV312nm

Sample: $Sc_2@C_{76}(I)$
$Sc_2@C_{78}$
$Sc_2@C_{80}(I)$
C_{86}

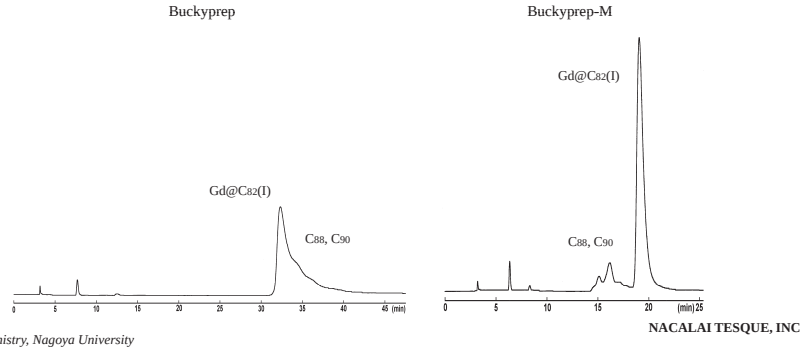


Sample courtesy of Prof. H.Shinohara,Department of chemistry, Nagoya University

COSMOSIL Application Data

Column:
Column size: 4.6mmI.D.-250mm
Mobile phase: Toluene
Flow rate: 1.0 ml/min
Temperature: 30°C
Detection: UV312nm

Sample: $Gd@C_{82}(I)$



Sample courtesy of Prof. H.Shinohara,Department of chemistry, Nagoya University

Ordering Information

- Analytical / Preparative Column (Particle Size: 5 µm)

COSMOSIL Buckyprep-M Packed Column

Column Size I.D. x Length (mm)	Product Number
4.6 x 250	04138-71
10 x 250	04141-11
20 x 250	04142-01
28 x 250	05873-01

COSMOSIL Buckyprep-M Guard Column

Column Size I.D. x Length (mm)	Product Number
4.6 x 10	04139-61
10 x 20	04140-21
20 x 50	34474-31
28 x 50	05872-11

COSMOSIL PBB

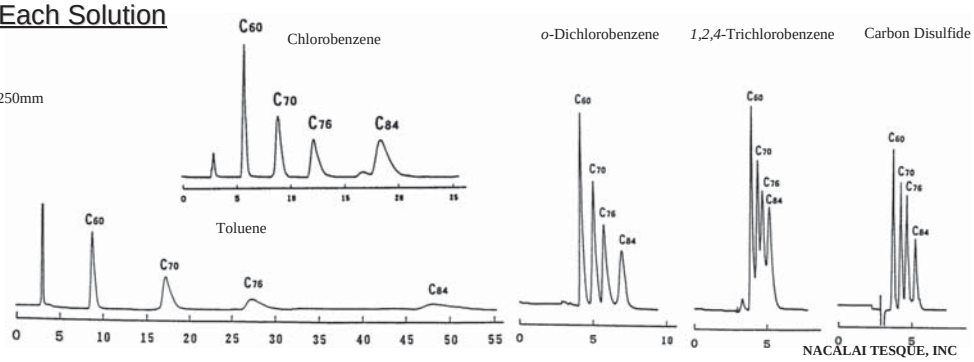
- Designed for preparative separation of fullerenes and higher fullerenes
- Can be used with *o*-dichlorobenzene or carbon disulfide
- Suitable for preparative scale separation

Separation of Fellerenes with Different Mobile Phases

Separation in Each Solution

Column: PBB
Column size: 4.6mmI.D.-250mm
Mobile phase:
Flow rate: 1.0 ml/min
Temperature: 30°C
Detection: UV285nm

Sample: C_{60} C_{70} C_{76} C_{84}



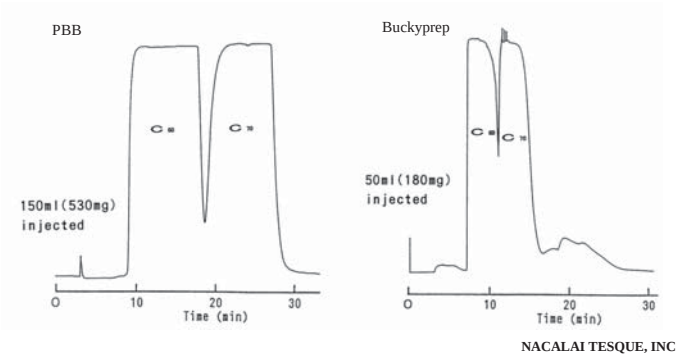
Preparative-scale Separation

The loading capacity of COSMOSIL PBB for C_{60} and C_{70} can be three times greater than COSMOSIL Buckyprep.

Preparative Separation of Fullerenes

Column:
Column size: 20mmI.D.-250mm
Mobile phase: Toluene
Flow rate: 18 ml/min
Temperature: Room temperature
Detection: UV285nm

Sample: Crude fullerenes (3.5mg/ml)



Ordering Information

● Analytical / Preparative Column (Particle Size: 5 µm)

COSMOSIL 5PBB Packed Column

Column Size I.D. x Length (mm)	Product Number
4.6 x 250	37980-01
10 x 250	37985-51
20 x 250	37986-41

COSMOSIL 5PBB Guard Column

Column Size I.D. x Length (mm)	Product Number
4.6 x 10	37987-31
10 x 20	37988-21
20 x 50	34375-31

COSMOSIL NPE

- Different selectivity from Buckyprep or PBB
- Excellent separation for derivatized fullerenes

Applications

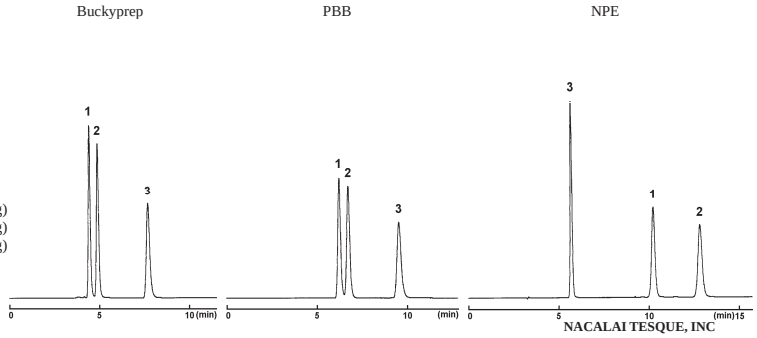
- PCBM, PCBB

COSMOSIL NPE retains derivatized C_{60} stronger than C_{60}.

COSMOSIL Application Data

Column:
Column size: 4.6mmI.D.-250mm
Mobile phase: (Buckyprep, PBB) Toluene
(NPE) Toluene/ Hexane = 25/75
Flow rate: 1.0 ml/min
Temperature: 30°C
Detection: UV325nm

Sample:
1; [6,6]-Phenyl-C_{61} Butyric Acid Methyl Ester [PCBM] (1.5µg)
2; [6,6]-Phenyl-C_{61} Butyric Acid Butyl Ester [PCBB] (1.5µg)
3; C_{60} (1.5µg)



Need to add the Hexane into mobile phase due to weak retention effect.

Ordering Information

- Analytical / Preparative Column (Particle Size: 5 µm)

COSMOSIL 5NPE Packed Column

Column Size I.D. x Length (mm)	Product Number
4.6 x 150	37902-21
4.6 x 250	37990-71
10 x 250	05469-11
20 x 250	38046-21

COSMOSIL 5NPE Guard Column

Column Size I.D. x Length (mm)	Product Number
4.6 x 10	37904-01
10 x 20	38045-31
20 x 50	05869-71

COSMOSIL PYE

Ordering Information

- Analytical / Preparative Column (Particle Size: 5 µm)

COSMOSIL 5PYE Packed Column

Column Size I.D. x Length (mm)	Product Number
4.6 x 250	37989-11
10 x 250	37996-11
20 x 250	38044-41
28 x 250	34300-91

COSMOSIL 5PYE Guard Column

Column Size I.D. x Length (mm)	Product Number
4.6 x 10	37903-11
10 x 20	38041-71
20 x 50	34475-21

11. Columns for Soluble Carbon Nanotube Separation

COSMOSIL CNT-300,CNT-1000,CNT-2000

- Size-based separation of soluble carbon nanotubes
- Three types of pore size (300 Å, 1000 Å, 2000 Å)
- High durability

Specifications

Packing Material	CNT-300	CNT-1000	CNT-2000
Silica Gel	High Purity Porous Spherical Silica		
Average Particle Size	5 µm		
Average Pore Size	approx. 300 Å	approx. 1000 Å	approx. 2000 Å
Bonded Phase	Hydrophilic Group (neutral)		
pH Range	2.0-7.5		
Pressure	15MPa and below		

Applications

- Carbon nanotubes

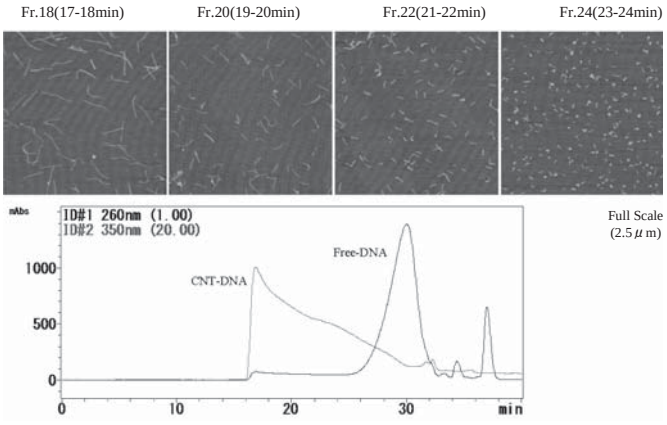
COSMOSIL CNT columns offered improved separation for DNA wrapped carbon nanotubes by connecting three columns with different pore sizes.

COSMOSIL Application Data

Column: CNT-300 + CNT-1000 + CNT-2000
Column size: 7.5mmI.D.-300mm × 3
Mobile phase: 0.5mmol/l EDTA, 200mmol/l NaCl, 40mmol/l Tris-Tris HCl(pH7.0)
Flow rate: 1.0 ml/min
Temperature: 40°C
Detection: UV260nm(× 1), 350nm(× 20)

Sample: DNA-wrapped CNT(CNT-DNA)
Inj. Vol. 500 µl

Data courtesy of Prof. H.Shinohara, Dr. Y.Asada, Department of chemistry, Nagoya University



AP-1147

Ordering Information

- Analytical column (Particle Size: 5µm)

COSMOSIL CNT-300 Packed Column

Column Size I.D. x Length (mm)	Product Number
7.5 x 300	09195-71

COSMOSIL CNT-300 Guard Column

Column Size I.D. x Length (mm)	Product Number
7.5 x 50	09194-81

COSMOSIL CNT-1000 Packed Column

Column Size I.D. x Length (mm)	Product Number
7.5 x 300	09197-51

COSMOSIL CNT-1000 Guard Column

Column Size I.D. x Length (mm)	Product Number
7.5 x 50	09196-61

COSMOSIL CNT-2000 Packed Column

Column Size I.D. x Length (mm)	Product Number
7.5 x 300	09199-31

COSMOSIL CNT-2000 Guard Column

Column Size I.D. x Length (mm)	Product Number
7.5 x 50	09198-41

12. Conventional Columns

Introduction

A period of more than 32 years has passed since the first COSMOSIL 5C$_{18}$ columns were developed and offered for sale. Continuous technical improvement has made many of these columns obsolete and not of the highest quality and performance available any more. However, many long-term users continue to employ these older conventional columns for routine analysis and quality control. Nevertheless, the manufacture of these older columns will eventually cease and we strongly urge customers to replace the conventional columns with their higher performance equivalents outlined in the table below. For additional information, contact the manufacturer or your local distributor directly.

Conventional Columns versus High Performance Columns

Conventional Columns versus High Performance Column List

Conventional Columns (old)		High Performance Columns (new)
5C$_{18}$-AR	→	5C$_{18}$-AR-II
5C$_{18}$	→	5C$_{18}$-MS-II
5C$_{18}$-MS	→	5C$_{18}$-MS-II
5C$_{18}$-P	→	5C$_{18}$-PAQ
5C$_{18}$-P-MS	→	5C$_{18}$-PAQ
5C$_{8}$	→	5C$_{8}$-MS
5TMS	→	5TMS-MS
5PE	→	5PE-MS
5CN-R	→	5CN-MS
5NH$_2$	→	5NH$_2$-MS
5C$_{18}$-300	→	5C$_{18}$-AR-300
5C$_{8}$-300	→	5C$_{8}$-AR-300
5C$_{4}$-300	→	5C$_{4}$-AR-300
5SL	→	5SL-II

For information on performance comparison of COSMOSIL 5C$_{18}$-MS-II (new), 5C$_{18}$-MS (old) and 5C$_{18}$ (old), please refer to page 213.
For COSMOSIL 5C$_{18}$-AR-II (new) and 5C$_{18}$-AR (old), please refer to page 214.

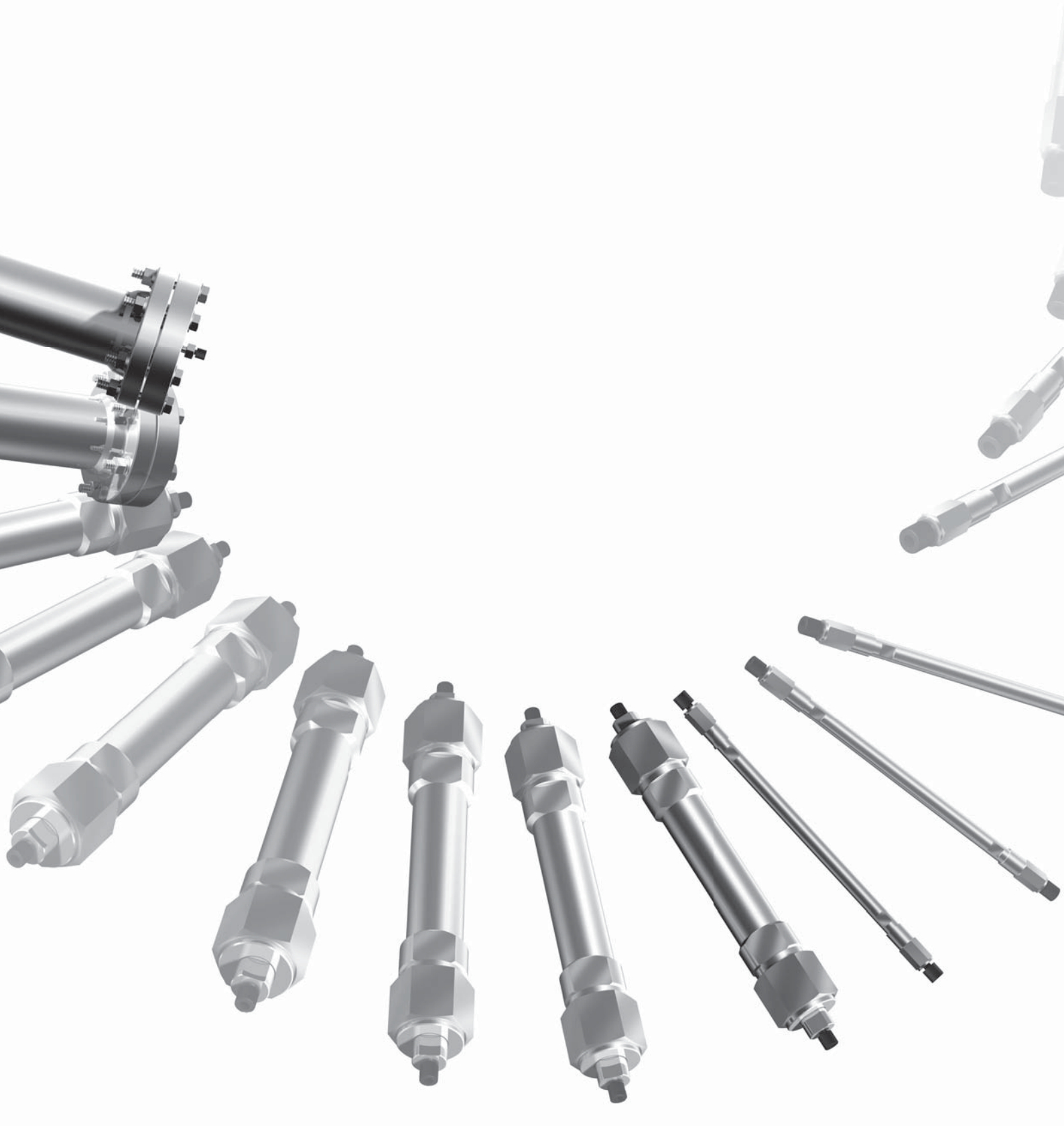
Ordering Information

Conventional Columns List

Product Name	Column Size I.D. x Length (mm)	Product Number
COSMOSIL 5C$_{18}$ Packed Column	4.6 x 150	39047-81
	4.6 x 250	39265-21
COSMOSIL 5C$_{18}$-MS Packed Column	4.6 x 150	37971-21
	4.6 x 250	37972-11
COSMOSIL 5C$_{18}$-AR Packed Column	4.6 x 150	37861-61
	4.6 x 250	37862-51
COSMOSIL 5C$_{18}$-P Packed Column	4.6 x 150	39103-31
	4.6 x 250	39280-11
COSMOSIL 5C$_{18}$-P-MS Packed Column	4.6 x 150	37995-21
	4.6 x 250	37994-31
COSMOSIL 5C$_{8}$ Packed Column	4.6 x 150	39042-31
	4.6 x 250	39260-71
COSMOSIL 5TMS Packed Column	4.6 x 150	39057-51
	4.6 x 250	39275-91
COSMOSIL 5CN-R Packed Column	4.6 x 150	39114-91
	4.6 x 250	39285-61
COSMOSIL 5NH$_2$ Packed Column	4.6 x 150	39150-11
	4.6 x 250	39290-81
COSMOSIL 5C$_{18}$-300 Packed Column	4.6 x 150	39607-41
COSMOSIL 5SL Packed Column	4.6 x 150	39037-11
	4.6 x 250	39255-51

Other conventional columns may available, please inquire.

II UHPLC Columns



COSMOSIL 2.5C_{18}-MS-II, 2.5Cholester, 2.5πNAP

- Low back pressure (2.5 μm silica gel)
- 3 types of stationary phases (monomeric C_{18}, cholesterol group and naphthalene group)

Specifications

Packing Material	C_{18}-MS-II	Cholester	πNAP
Silica Gel	High Purity Porous Spherical Silica		
Average Particle Size	2.5 μm		
Average Pore Size	aprrox. 130 Å		
Specific Surface Area	aprrox. 330 m^2/g		
Bonded Phase	Octadecyl Group	Cholesteryl Group	Naphtylethyl Group
Bonding Type	Monomeric Type		
Main Interaction	Hydrophobic Interaction	Hydrophobic Interaction Molecular Shape Selectivity	Hydrophobic Interaction π-π Interaction
End-capping Treatment	Near-perfect Treatment		
Carbon Load	aprrox. 18%	aprrox. 21%	aprrox. 14%
Feature	・First choice of reversed phase column	・The same mobile phase as C_{18} ・High molecular shape selectivity	・Stronger π-π interactions than phenyl columns

Ultra High Performance Liquid Chromatography (UHPLC)

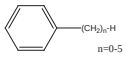
Very fast and efficient separation can be achieved using 2.5 μm particles.

Note: Ultra high performance liquid chromatography system or some modification of HPLC system is required for UHPLC analysis. The following application is acquired by HPLC equipment for semi-micro column, and response of detector is 0.02 sec.

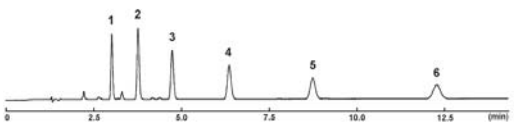
To Ultra Fast Liquid Chromatography

Column: COSMOSIL C_{18}-MS-II
Column size:
Mobile phase: Acetonitrile/ H_2O = 70/30
Flow rate: 1.0 ml/min
Temperature: 40°C
Detection: UV254nm

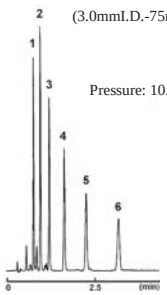
Sample:		
	1; Benzene	(1.67mg/ml)
	2; Toluene	(1.67mg/ml)
	3; Ethylbenzene	(1.67mg/ml)
	4; *n*-Propylbenzene	(1.67mg/ml)
	5; *n*-Butylbenzene	(1.67mg/ml)
	6; Amylbenzene	(1.67mg/ml)
Injection Vol.	1.0 μl	



$5C_{18}$-MS-II
(4.6mmI.D.-150mm)
Pressure: 3.7MPa



$2.5C_{18}$-MS-II
(3.0mmI.D.-75mm)
Pressure: 10.9MPa



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Comparison of Analytical Pressure

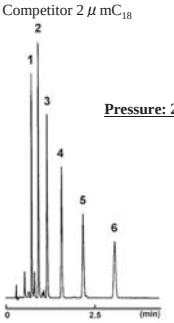
COSMOSIL 2.5 series can be used under lower pressure than competitors' 2 μm columns.

Comparison of Analytical Pressure

Column:
Column size: 3.0mmI.D.-75mm
Mobile phase: Acetonitrile/ H_2O = 70/30
Flow rate: 1.0 ml/min
Temperature: 40°C
Detection: UV254nm

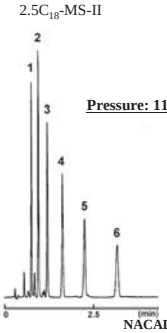
Sample:		
	1; Benzene	(1.67mg/ml)
	2; Toluene	(1.67mg/ml)
	3; Ethylbenzene	(1.67mg/ml)
	4; Propylbenzene	(1.67mg/ml)
	5; Butylbenzene	(1.67mg/ml)
	6; Amylbenzene	(1.67mg/ml)
Injection Vol.	1.0 μl	

Competitor 2 μm C_{18}



Pressure: 20.0MPa

$2.5C_{18}$-MS-II



Pressure: 11.0MPa

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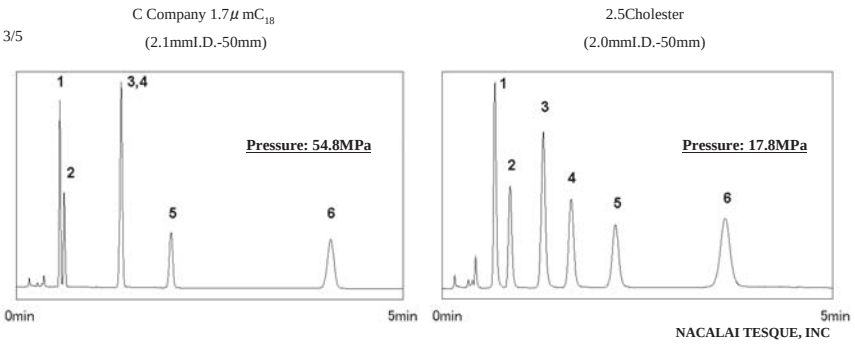
Applications

COSMOSIL 2.5Cholester offers improved resolution for compounds difficult to analyze with C_{18} without changing analytical condition. For more information on Cholester, refer to page 21.

● Saikosaponins

COSMOSIL Application Data

Mobile phase: Acetonitrile/ 0.05%NaH_2PO_4aq. = 3/5
Flow rate: 0.7 ml/min
Temperature: 50°C
Detection: UV206nm
Sample: 1; Saikosaponin c
2; Saikosaponin h
3; Saikosaponin a
4; Saikosaponin b2
5; Saikosaponin b1
6; Saikosaponin d
Injection Vol. 1.0 μl



Data courtesy of a customer

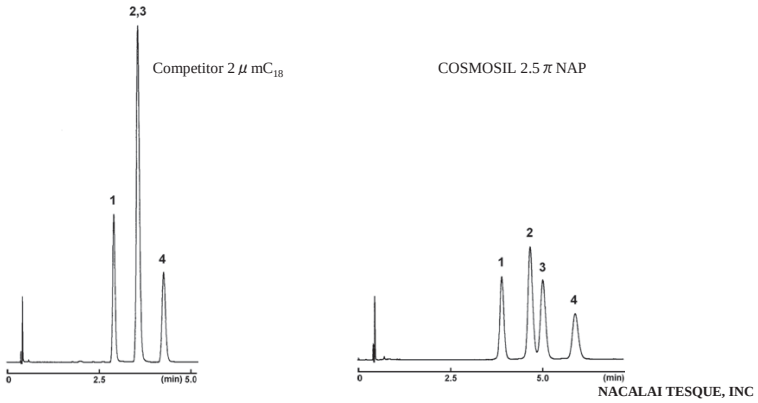
COSMOSIL 2.5πNAP provides greater performance in separating positional isomers and other closely related compounds which are difficult to analyze with C_{18}. For more information on πNAP, refer to page 24.

● Tocopherols

COSMOSIL Application Data

Column:
Column size: 3.0mmI.D.-75mm
Mobile phase: Competitor 2 μ mC_{18} Methanol / H_2O = 95/5
2.5 π NAP Methanol / H_2O = 85/15
Flow rate: 1.0 ml/min
Temperature: 40°C
Detection: UV295nm

Sample: 1; δ -Tocopherol
2; γ -Tocopherol
3; β -Tocopherol
4; α -Tocopherol



AP-1151

Orderling Information

● Analytical Column (Particle Size: 2.5 μm)

COSMOSIL 2.5C_{18}-MS-II Packed Column

Column Size I.D. x Length (mm)	Product Number
2.0 x 50	08994-31
2.0 x 75	08995-21
2.0 x100	08996-11
3.0 x 50	08997-01
3.0 x 75	08998-91
3.0 x100	08999-81

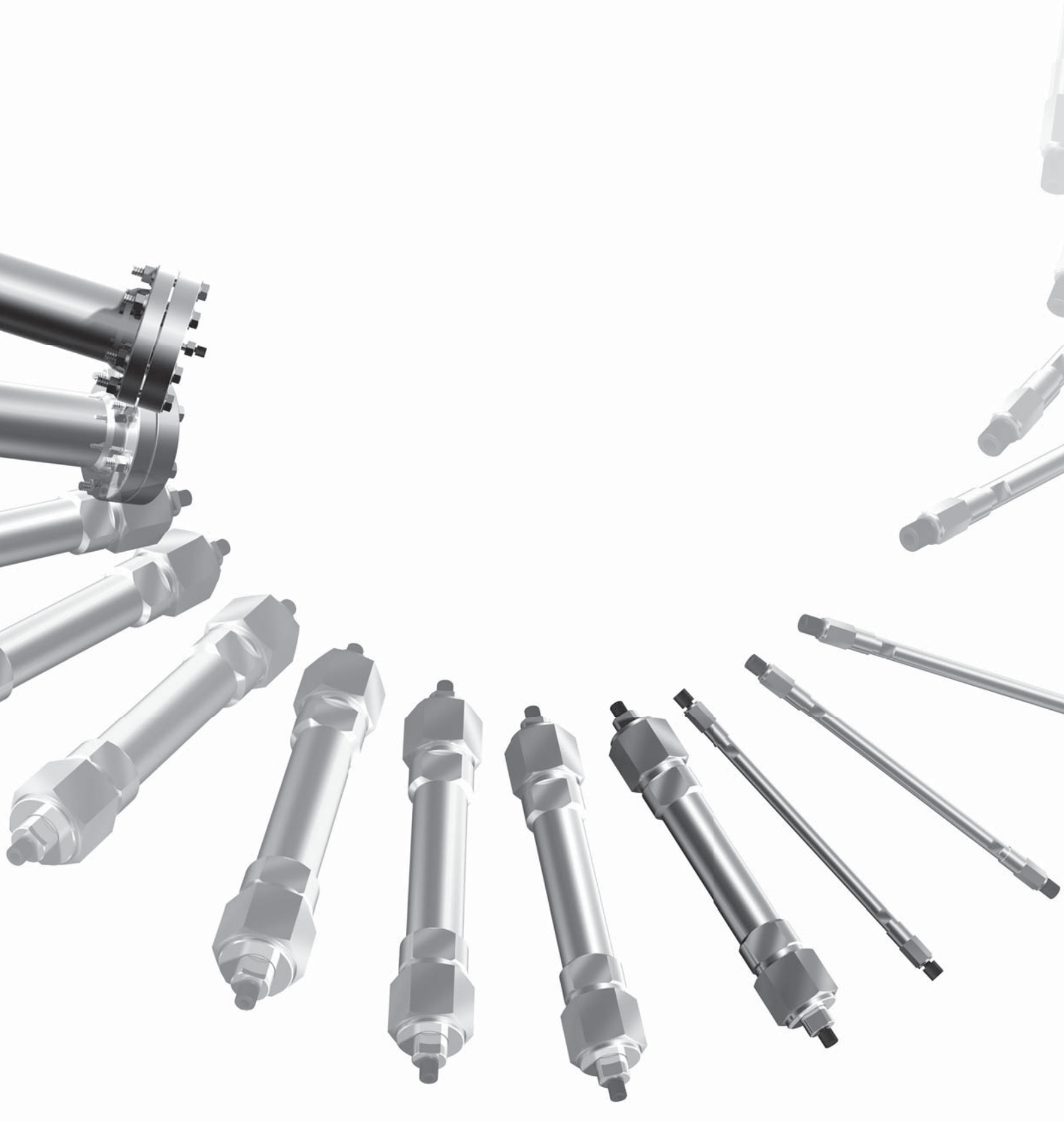
COSMOSIL 2.5Cholester Packed Column

Column Size I.D. x Length (mm)	Product Number
2.0 x 50	09000-01
2.0 x 75	09047-11
2.0 x 100	09048-01
3.0 x 50	09049-91
3.0 x 75	09050-51
3.0 x 100	09051-41

COSMOSIL 2.5πNAP Packed Column

Column Size I.D. x Length (mm)	Product Number
2.0 x 50	06062-91
2.0 x 75	06051-31
2.0 x 100	06052-21
3.0 x 50	06054-01
3.0 x 75	06055-91
3.0 x 100	06057-71

III Preparative Packing Materials



1. Normal and Reversed Phase Packing Materials

Introduction

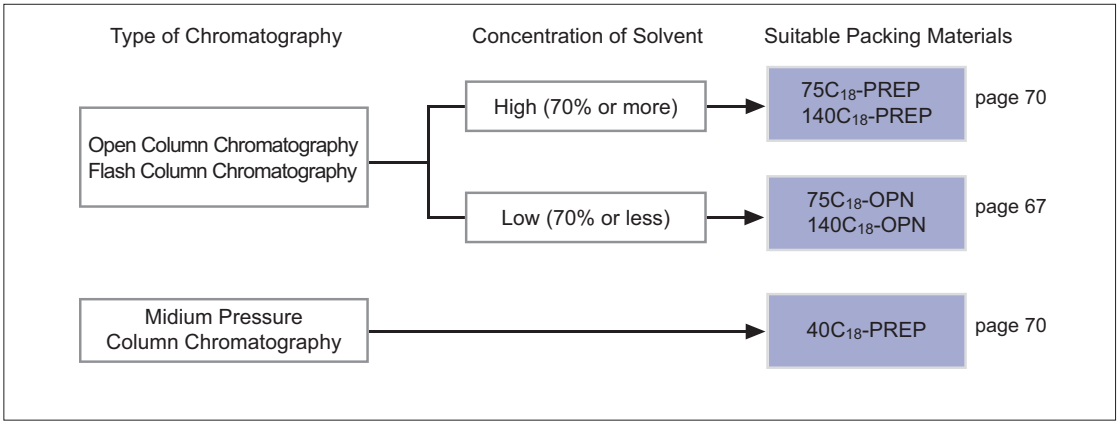
Open column chromatography is an excellent and easy technique for large-scale preparation and purification at low cost. COSMOSIL offers both normal and reversed phase packing materials based on totally porous spherical silica, which provides higher separation, less pressure and higher reproducibility than irregular silica.

Specifications

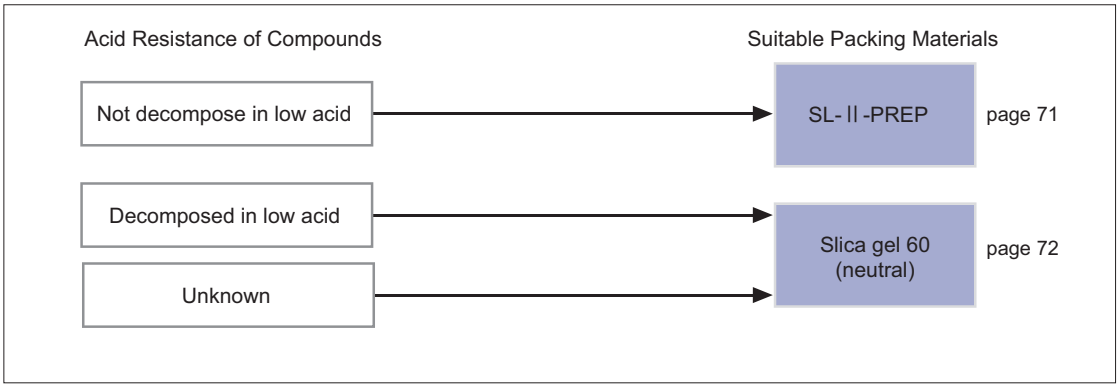
Packing Material	C_{18}-OPN	C_{18}-PREP	SL-II-PREP	Silica Gel 60 (neutral)
Silica Gel	High Purity Porous Spherical Silica			
Average Particle Size	75, 140 µm	40, 75, 140 µm	75, 140 µm	
Average Pore Size	approx.120 Å			approx. 60 Å
Specific Surface Area	approx. 300 m^2/g			approx. 500 m^2/g
Bonded Phase	Octadecyl Group		None	
Carbon Load	—	approx. 19%	0%	
Residual Silanol Group	Yes	None	—	
Application	Open Column Chromatography / Flash Column Chromatography			
	Reversed Phase Chromatography		Normal Phase Chromatography	

For more informations on other silica gel, please refer to page 73.

Selection Guide (reversed phase)



Selection Guide (normal phase)



COSMOSIL C_{18}-OPN

- A new “Water-Wet” C_{18} packing material for reversed phase open column chromatography
- Usable under 100% aqueous eluents

Characteristic

The external surface of the C_{18}-OPN gel is coated with hydrophilic group to increase wettability of the gel, and octadecyl group is bonded in the pore of the gel. Conventional reversed phase C_{18} packing materials are restricted to about 30-50% water in the mobile phase. The COSMOSIL C_{18}-OPN is a new “Water-Wet” C_{18} packing material developed for reversed phase open column chromatography. The C_{18}-OPN material can be used in 100% aqueous eluents.

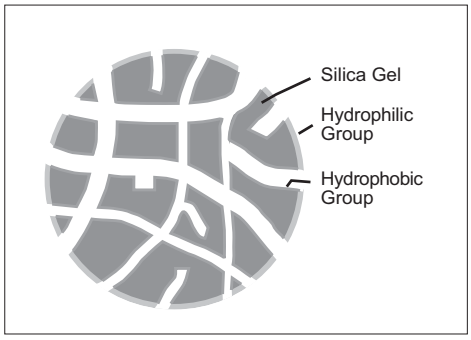


Figure 1. Structure of C_{18}-OPN

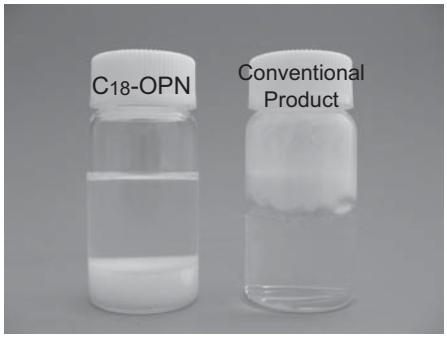


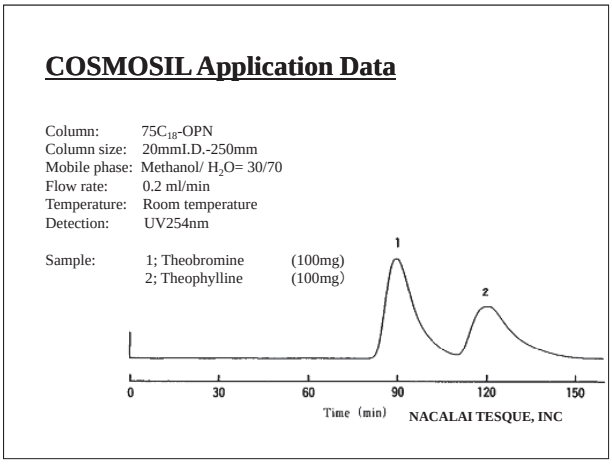
Figure 2. Packing material in water

Left: C_{18}-OPN provides good resolution
Can be used with low concentration of organic solvent on open, flash column chromatography.

Right: C_{18}-PREP float up
Use with 70% or more organic solvent on open, flash column chromatography.

Applications

- Separation of hydrophilic compounds in aqueous solution



In reversed phase chromatography, hydrophilic compounds such as Theobromine and Theophylline could be separated under low concentration of organic solvent. The figure shows they are clearly separated by reversed open column chromatography with 70% of water.

Influence of Particle Size

The table below shows comparison between 75 µm and 140 µm particle size silica. Although peak shapes and flow rate may differ based on particle size of silica, elution behavior is the same.

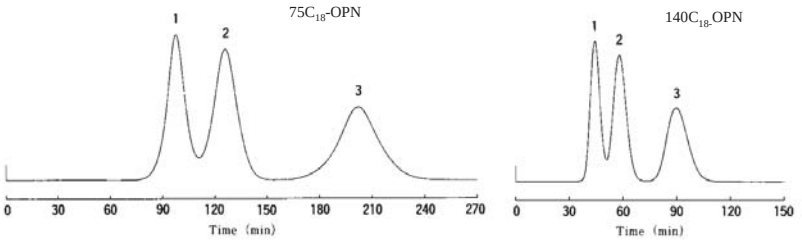
COSMOSIL Application Data

Influence of Particle Size

Particle	Flow Rate	Theoretical Plate Number			Rs		Separation Time	Solvent Consumption
(µm)	(ml/min)	TB	TP	CF	TB/TP	TP/CF	(min)	(ml)
75	0.25	400	390	340	1	1.74	240	60
140	0.6	300	280	260	0.9	1.4	100	60

Column:
Column size: 10mmI.D.-250mm
Mobile phase: Methanol/ H_2O= 20/80
Flow rate: 0.2 ml/min
Temperature: Room temperature
Detection: UV254nm

Sample: 1; Theobromine (TB)
2; Theophylline (TP)
3; Caffeine (CF)



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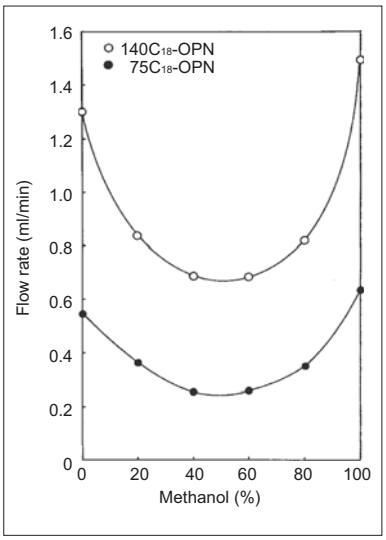
Flow Rate



Since reversed phase chromatography generally employs high viscosity solvents such as water and methanol, the flow rate is lower than that of normal phase chromatography. The flow rate of reversed phase depends on the mobile phase composition. The figure left indicates that the flow rate of the COSMOSIL 140C_{18}-OPN (140 µm in particle size) is about 2.5 times higher than that of the COSMOSIL 75C_{18}-OPN.

Figure. Concentration of methanol against flow rate
Column size: 10 mm I.D. x 180 mm bed height (gravitational liquid flow)

Comparison of Normal Phase

● Separation of *p*-Cresol and *p*-Ethylphenol by normal and reversed phase mode

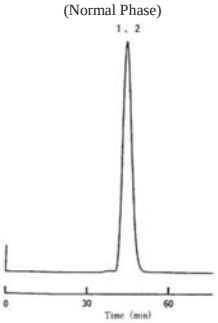
Since the structural difference between *p*-Cresol and *p*-Ethylphenol is only one methyene group, it is difficult to separate such samples under normal phase condition. On the other hand, the samples are clearly separated under reversed phase condition with COSMOSIL C_{18}-OPN packing material.

COSMOSIL Application Data

(Normal Phase)
Packing media: Silica Gel 120(150~325mesh)
Column size: 10mmI.D.-180mm
Mobile phase: Hexane/Ethyl Acetate = 90/10
Flow rate: 0.7 ml/min
Temperature: room temperature
Detection: UV254nm

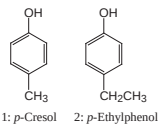
Sample: 1; *p*-Cresol
2; *p*-Ethylphenol

(Normal Phase)



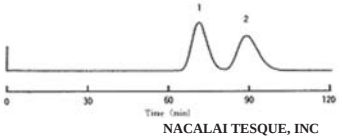
(Reversed Phase)
Packing media: 75C_{18}-OPN
Column size: 10mmI.D.-180mm
Mobile phase: Methanol/H_2O = 60/40
Flow rate: 0.2ml/min
Temperature: room temperature
Detection: UV254nm

Sample: 1; *p*-Cresol
2; *p*-Ethylphenol



1: *p*-Cresol 2: *p*-Ethylphenol

(Reversed Phase)



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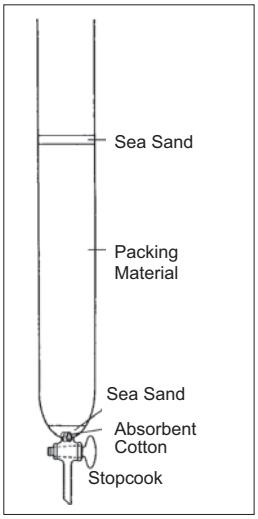
Column Packing Instructions



1. Use a standard open glass column, close the stopcock, pack a small amount of absorbent cotton in the bottom of the column and add solvent to approximately 1/3 of the column length.
2. Add a thin layer (5 mm) of sea sand to the surface of the absorbent cotton.
3. Prepare a slurry solution of the packing material (30% w/v) with solvent right before packing. (Make sure to prepare enough slurry solution to form a column bed sufficient to separate the compounds of interest.)
4. Simultaneously open the stopcock and add the slurry solution to the column to form the column bed.
5. After packing the column, wash the newly packed column bed with 5–10 column volumes of solvent. Allow the bed to stabilize overnight in solvent.
6. Add a thin layer (5 mm) of sea sand to the top of the bed in order to prevent disturbance of the top of the column bed during sample or solvent addition.

Column Size and Required Amount of Packing Material

Table. Column size and required amount of C_{18}-OPN packing material

Column I.D. (mm)	Bed Height (mm)	Amount of C_{18}-OPN (g)
10	150	4
	250	7
20	150	17
	250	28
30	150	38
	250	63

Reproducibility and Washing Methods

Wash the COSMOSIL C_{18}-OPN packing material with tetrahydrofuran, chloroform or other solvents to remove the impurities. This packing material has excellent reproducibility and can be used repeatedly.

"Attention"

1. Do not wash with basic solvents of pH 7 or more which will dissolve the silica gel or pH 2 or less which will cleave the C_{18} stationary phase.
2. Dry the packing material at 50°C or less.

Ordering Information

● COSMOSIL C_{18}-OPN

Product Name	Average Particle Size	Product Number	PKG Size
COSMOSIL 75C_{18}-OPN	75µm	37842-66	100 g
		37842-95	500 g
		37842-11	1 kg
COSMOSIL 140C_{18}-OPN	140µm	37878-16	100 g
		37878-45	500 g
		37878-61	1 kg

COSMOSIL C_{18}-PREP

- Standard reversed phase packing material for open chromatography
- End-capping treated
- 3 types of particle size (40, 75, 140 µm)

Particle Size, Flow Rate and Theoretical Plate Number

Because reversed phase chromatography employs mobile phase of high viscosity such as methanol and water, the flow rate is lower than that of normal phase chromatography, which uses mobile phase of low viscosity such as hexane and ethyl acetate.

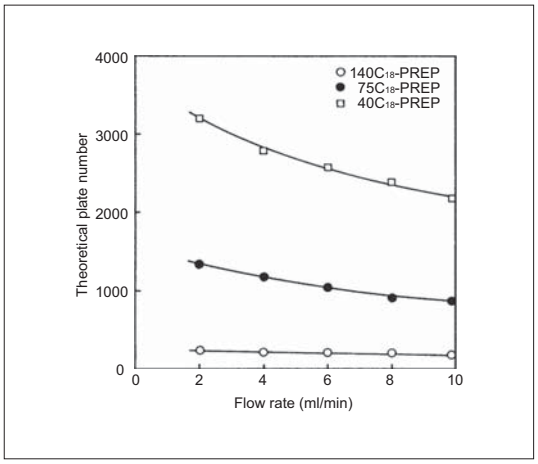


Figure 1. Flow rate against theoretical plate number

Column size: 20 mm I.D. x 300 mm

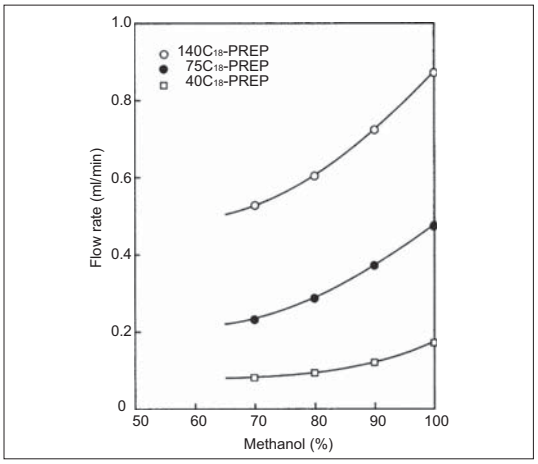


Figure 2. Concentration of methanol against flow rate

Column size: 10 mm I.D. x 180 mm bed height
(gravitational liquid flow)

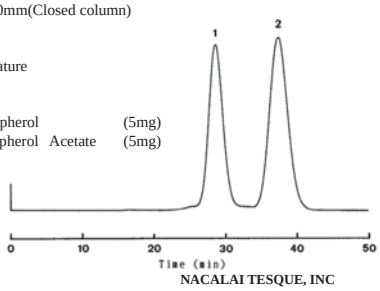
Applications

- Vitamin E

COSMOSIL Application Data

Column: 40C_{18}-PREP
Column size: 20mmI.D.-300mm(Closed column)
Mobile phase: Methanol
Flow rate: 9.9 ml/min
Temperature: Room temperature
Detection: UV280nm

Sample: 1; *DL*-α-Tocopherol (5mg)
2; *DL*-α-Tocopherol Acetate (5mg)



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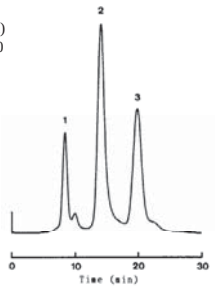
- Natural Compounds

COSMOSIL Application Data

Column: 40C_{18}-PREP
Column size: 20mmI.D.-300mm(Closed column)
Mobile phase: Methanol/ 0.05%TFA-H_2O = 70/30
Flow rate: 9.9 ml/min
Temperature: Room temperature
Detection: UV254nm

Sample: 1; Baicalin (40µg)
2; Baicalein (120µg)
3; Wogonin (40µg)



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Ordering Information

- COSMOSIL C_{18}-PREP

Product Name	Average Particle Size	Product Number	PKG Size
COSMOSIL 40C_{18}-PREP	40 µm	37932-86	100 g
		37932-15	500 g
		37932-31	1 kg
COSMOSIL 75C_{18}-PREP	75 µm	37933-76	100 g
		37933-05	500 g
		37933-21	1 kg
COSMOSIL 140C_{18}-PREP	140 µm	37934-66	100 g
		37934-95	500 g
		37934-11	1 kg

COSMOSIL SL-II-PREP

- Standard packing materials for normal phase chromatography
- Ultra pure silica gel packing material more than 99.99% purity

Performance for Chelating Compounds

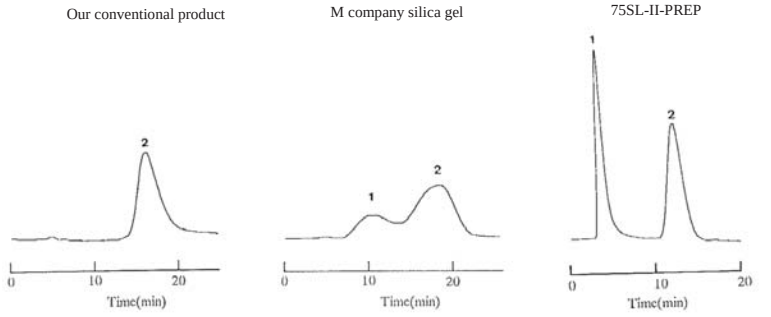
Highly purified silica gel of COSMOSIL SL-II-PREP enables separation of metal coordination compounds without adsorption.

- Metal Coordination Compounds

COSMOSIL Application Data

Column:
Column size: 10mmI.D.-250mm
Mobile phase: Hexane/Ethanol = 95/5
Flow rate: 5.0 ml/min
Temperature: 30°C
Detection: UV254nm

Sample: 1; Quinizarin
2; *p*-Nitrobenzyl Alcohol



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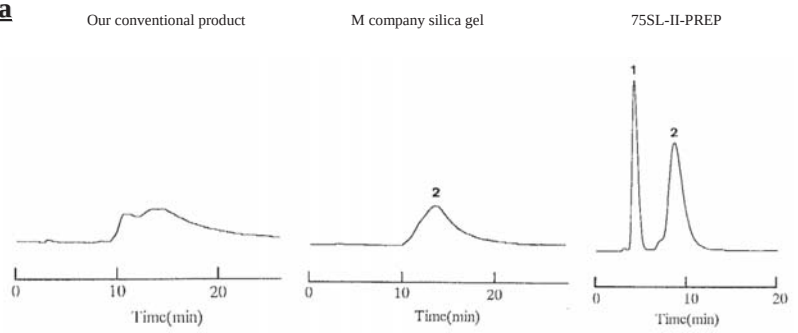
- Organic Acid and Amide

COSMOSIL Application Data

Column:
Column size: 10mmI.D.-250mm
Mobile phase: Hexane/Ethanol = 90/10
Flow rate: 5.0 ml/min
Temperature: 30°C
Detection: UV254nm

Sample: 1; Salicylic Acid
2; Salicylamide



NACALAI TESQUE, INC

Ordering Information

- COSMOSIL SL-II-PREP

Product Name	Average Particle Size	Product Number	PKG Size
COSMOSIL 75SL-II-PREP	75 µm	38012-64	100 g
		38012-35	500 g
		38012-51	1 kg
COSMOSIL 140SL-II-PREP	140 µm	38013-54	100 g
		38013-41	1 kg

Silica Gel (spherical, neutral)

- The pH of Silica Gel is adjusted to neutral
- Suitable for the separation of pH sensitive compounds

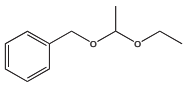
Comparison with Conventional Silica Gel

- Purification of Acetal -1

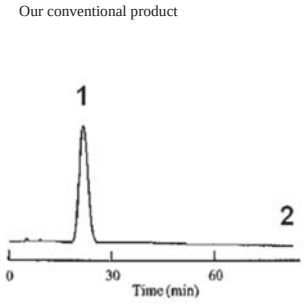
COSMOSIL Application Data

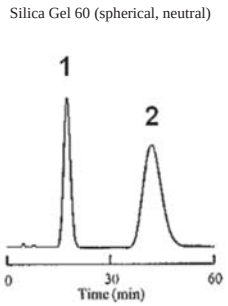
Column:
Column size: 4.6mmI.D.-250mm
Mobile phase: Hexane/Ethyl Acetate = 99/1
Flow rate: 1.0 ml/min
Temperature: 30°C
Detection: UV254nm

Sample: 1; Methyl Benzoate(Standard) (10mg/ml)
2; Sample A (100mg/ml)
Inj.Vol. 3 μl



Sample A





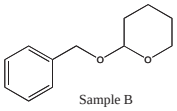
NACALAI TESQUE, INC

- Purification of Acetal -2

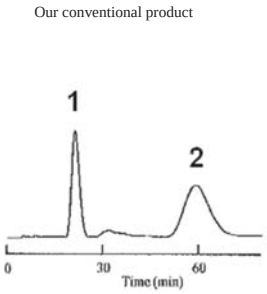
COSMOSIL Application Data

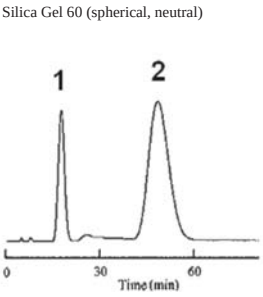
Column:
Column size: 4.6mmI.D.-250mm
Mobile phase: Hexane/Ethyl Acetate = 99/1
Flow rate: 1.0 ml/min
Temperature: 30°C
Detection: UV254nm

Sample: 1; Methyl Benzoate(Standard) (10mg/ml)
2; Sample B (200mg/ml)
Inj.Vol. 3 μl



Sample B





NACALAI TESQUE, INC

Ordering Information

- Silica gel 60 (spherical, neutral)

Product Name	Average Particle Size	Product Number	PKG Size
Silica Gel 60 (spherical, neutral) for Column Chromatograph	75 μm	30511-64	100 g
		30511-35	500 g
		30511-51	1 kg
		30511-06	5 kg
		30511-22	25 kg
	140 μm	30518-94	100 g
		30518-65	500 g
		30518-81	1 kg
		30518-52	25 kg

Silica Gel (for column chromatograph)

Ordering Information

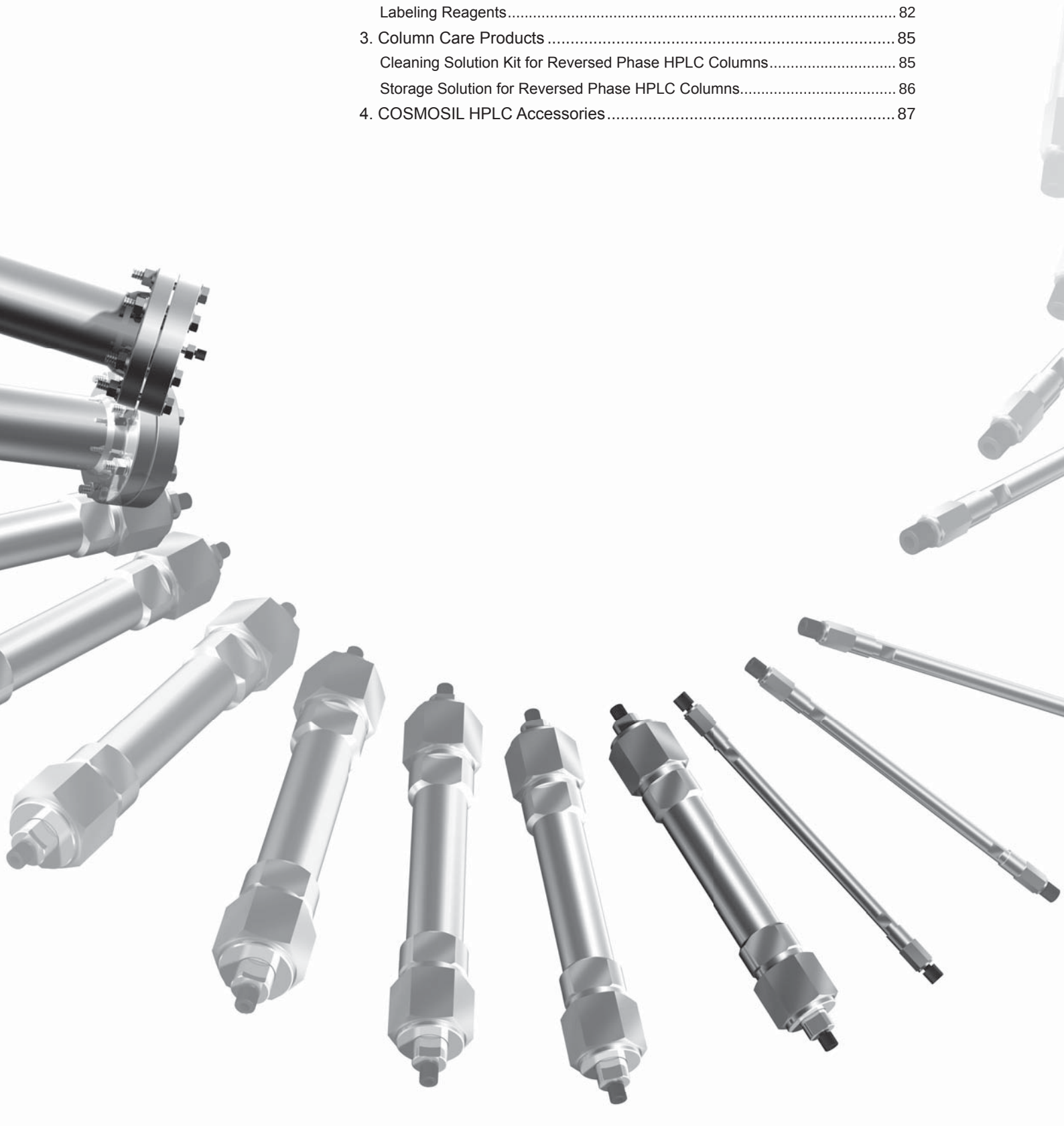
● Silica Gel (spherical)

Product Name	Particle Size	Pore Size	Grade	Product Number	PKG Size
Silica Gel 60, Spherical	approx. 70 ~ 230 mesh	60 Å	SP	30731-71	1 kg
				30731-42	25 kg
	approx. 150 ~ 325 mesh		SP	30733-51	1 kg
				30733-22	25 kg
Silica Gel 120, Spherical	approx. 70 ~ 230 mesh	120 Å	SP	30734-41	1 kg

● Silica Gel (irregular)

Product Name	Particle Size	Pore Size	Grade	Product Number	PKG Size
Silica Gel 60	approx. 70 ~ 230 mesh	60 Å	SP	30724-55	500 g
				30724-71	1 kg
				30724-84	5 kg
				30724-42	25 kg
	approx. 230 ~ 400 mesh		SP	30721-85	500 g
				30721-01	1 kg
				30721-14	5 kg

IV Related Products



1. Reagents for Mobile Phase Preparation

Solvents, Additives

Specification

HPLC grade solvents and additives are specifically designed for HPLC applications with the specifications including; purity, specific gravity, refraction index, absorbance, water, non-volatile matter, peroxide and fluorescence.

* Specifications vary by solvent.

Ordering Information

● Solvents

Product Name	Grade	Product Number	Package Size
Acetone	SP	00325-31	1 L
Acetonitrile	SP	00430-25	500 ml
		00430-41	1 L
		00430-83	3 L
Benzene	SP	04028-11	1 L
1-Butanol	SP	06024-91	1 L
t-Butyl Methyl Ether	SP	06332-64	200 ml
		06332-51	1 L
Chloroform	SP	08426-71	1 L
		08426-13	3 L
Cyclohexane	SP	10034-31	1 L
o-Dichlorobenzene	SP	11635-31	1 L
1,2-Dichloroethane	SP	15223-01	1 L
Dichloromethane	SP	22423-61	1 L
N,N-Dimethylformamide	SP	13024-71	1 L
1,4-Dioxane	SP	13631-11	1 L
Distilled Water	SP	14029-91	1 L
		14029-33	3 L
Ethanol (99.5V%)	SP	14741-25	500 ml
		14741-41	1 L
		14741-83	3 L
Ethyl Acetate	SP	14746-91	1 L
		14746-33	3 L
Heptane	SP	17623-01	1 L
1,1,1,3,3,3-Hexafluoro-*2*-propanol	SP	17814-14	100 g
		17814-85	500 g
Hexane	SP	17929-11	1 L
		17929-53	3 L
Methanol	SP	21929-81	1 L
		21929-23	3 L
Phosphate Buffer Solution (pH 2.5) (5x)	SP	08969-71	1 L
Phosphate Buffer Solution (pH 7.0) (5x)	SP	08968-81	1 L
1-Propanol	SP	29033-61	1 L
2-Propanol	SP	29128-31	1 L
		29128-73	3 L
Tetrahydrofuran	SP	33125-31	1 L
		33125-73	3 L
Toluene	SP	34130-21	1 L
		34130-63	3 L

● Additives

Product name	Grade	Product Number	Package Size
Acetic Acid	SP	08963-02	25 ml
Formic Acid	SP	08965-82	25 ml
Phosphoric Acid, Ortho	SP	08964-92	25 ml
Trifluoroacetic Acid	SP	34840-21	5×1 ml
		34840-76	5×1.5 ml
		34840-63	5×3 ml
		34840-34	10 ml

Phosphate Buffer Solution (pH 2.5) (5x)

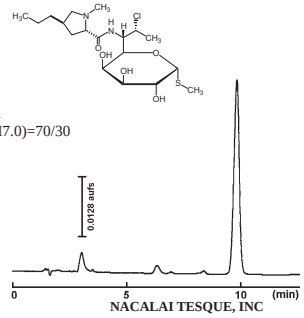
- pH adjusted
- Filtered (0.2 μm)
- UV, Fluorescence tested
- Easy to prepare the mobile phase used in COSMOSIL Applications

How to Prepare

Dilute this product 1:4 with HPLC grade distillated water to make the 20 mmol/l phosphate buffer used in the following COSMOSIL Applications.

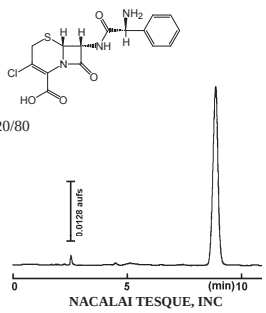
COSMOSIL Chromatogram Index

Sample:	Clindamycin
CAS No.:	[18323-44-9]
Molecular formula:	$C_{18}H_{33}ClN_2O_5S$
Column:	$5C_{18}$-MS-II
Column size:	4.6mmI.D.-150mm
Mobile phase:	Methanol/ 20mmol/l Phosphate buffer(pH7.0)=70/30
Flow rate:	1.0 ml/min
Temperature:	30°C
Detection:	UV210 nm
Attenuation:	0.128 aufs
Sample conc.:	1.0mg/ml
Injection volume:	3.0μl
Retention time:	9.86min
Capacity factor:	4.99



COSMOSIL Chromatogram Index

Sample:	Cefaclor
CAS No.:	[53994-73-3]
Molecular formula:	$C_{15}H_{14}ClN_3O_4S$
Column:	$5C_{18}$-MS-II
Column size:	4.6mmI.D.-150mm
Mobile phase:	Methanol/ 20mmol/l Phosphate buffer(pH2.5)=20/80
Flow rate:	1.0 ml/min
Temperature:	30°C
Detection:	UV220 nm
Attenuation:	0.128 aufs
Sample conc.:	1.0mg/ml
Injection volume:	0.5μl
Retention time:	8.93min
Capacity factor:	3.75



For more information on COSMOSIL Chromatogram Applications, please refer to page 90.

Attention

1. This product is designed to prepare the 20 mmol/l phosphate buffer. If the product is diluted to different concentrations, confirm pH before use.
2. Use distilled water (Product No. 14029).
3. Keep refrigerated and use promptly after opening.

Ordering Information

- Phosphate Buffer Solution (5x)

Product Name	Grade	Product Number	PKG Size
Phosphate Buffer Solution (pH 2.5) (5x)	SP	08969-71	1 L
Phosphate Buffer Solution (pH 7.0) (5x)	SP	08968-81	1 L

Ion-pair Reagents

Introduction

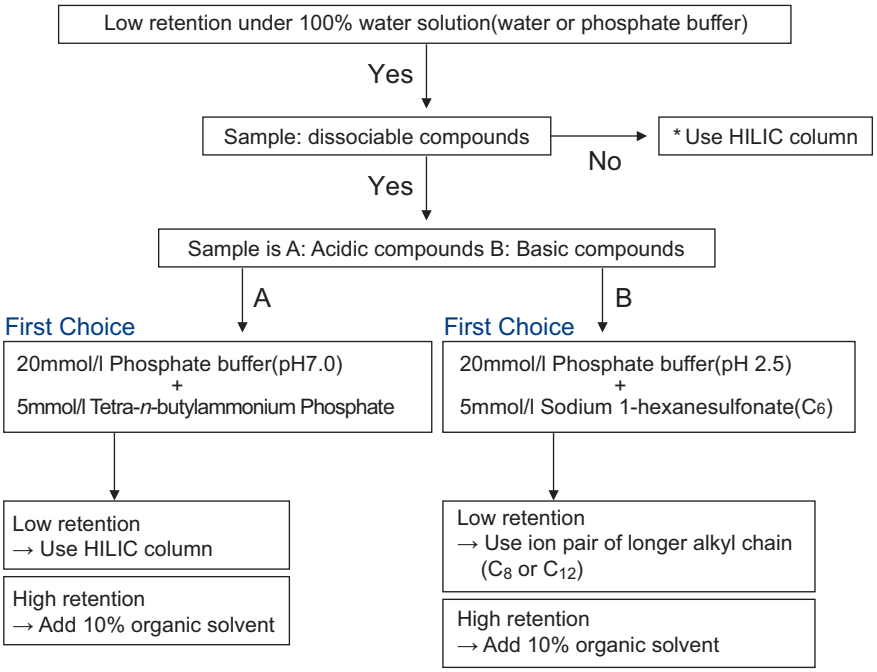
The use of ion pair reagents as mobile phase additives extends the applicability of reversed phase HPLC. Ionic or highly polar compounds are difficult to analyze by reversed phase using only organic solvent and buffer solution because of the short retention time. Ion pair reagents are strong hydrophobic ions which form neutral ion pairs with oppositely charged samples molecules, making the efficient ODS columns amenable to separate ionic or highly polar samples. Nacalai Tesque offers a broad range of ion pair reagents for pharmaceutical compounds and other highly polar materials.

General Use of Ion-pair Reagents

When using ion pair reagents, ample time should be allowed for establishing equilibrium and for cleaning the column.
When using ion pair reagents with an alkyl chain of C_{10} or shorter, it typically takes 20 minutes for establishing equilibrium and 30 minutes for cleaning. It may take more than 1 hour to clean the column when using ion pair reagents with an alkyl chain longer than C_{10}. Therefore, it is highly recommended to prepare a column for exclusive use with ion pair reagents.

Condition Setting

Follow the condition setting below.

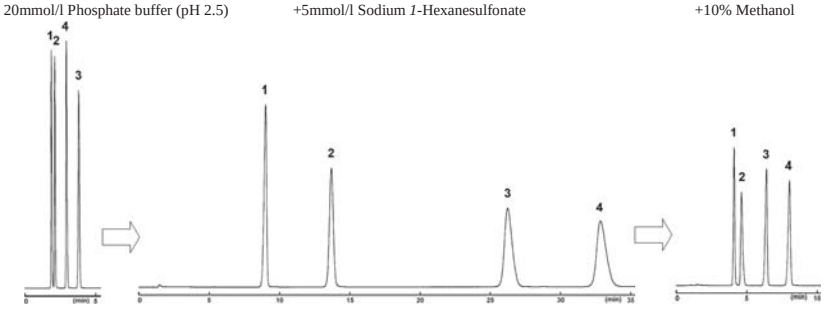


*For HILIC column, please refer to page 36.

Condition Setting

Column: $5C_{18}$-PAQ
Column size: 4.6mmI.D.-150mm
Mobile phase:
Flow rate: 1.0 ml/min
Temperature: 30°C
Detection: UV270nm

Sample: 1; *L*-Noradrenaline
2; *L*-Adrenaline
3; *L*-DOPA
4; Dopamine



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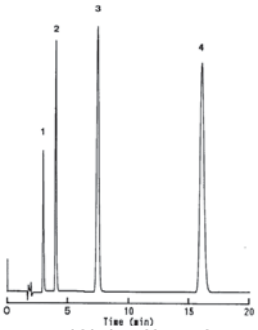
Applications

● Low-molecular-weight Unsaturated Carboxylic Acids

COSMOSIL Application Data

Column: $5C_{18}$-MS-II
Column size: 4.6mmI.D.-150mm
Mobile phase: Acetonitrile/ 5mmol/l Tetra-*n*-butylammonium Phosphate, 20mmol/l Phosphate buffer(pH7) = 10 /90
Flow rate: 1.0 ml/min
Temperature: 30°C
Detection: UV230nm, 0.5AUFS

Sample:
1; Acrylic Acid (2.0μg)
2; Crotonic Acid (2.0μg)
3; Tiglic Acid (2.0μg)
4; Sorbic Acid (2.0μg)



NACALAI TESQUE, INC

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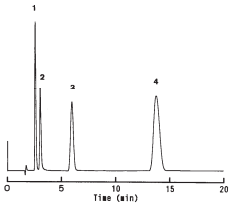
● Amino Acids

COSMOSIL Application Data

Column: $5C_{18}$-MS-II
Column size: 4.6mmI.D.-150mm
Mobile phase: Acetonitrile/ 5mmol/l Sodium *l*-Hexanesulfonate, 20mmol/l Phosphate buffer(pH3) = 10/90
Flow rate: 1.0 ml/min
Temperature: 30°C
Detection: UV215nm, 0.5AUFS

Sample:
1; *L*-Histidine (1.0μg)
2; *L*- Tyrosine (3.0μg)
3; *L*-(-)-Phenylalanine (1.0μg)
4; *L*- Tryptophan (0.5μg)



NACALAI TESQUE, INC

AP-0437

Ordering Information

● for Basic Samples ($R\text{-}SO_3^-Na^+$)

Product Name	R:	Grade	Product Number	PKG Size
Sodium *1*-Butanesulfonate	C_4H_9-	SP	31331-94	5 g
Sodium *1*-Pentanesulfonate	C_5H_{11}-	SP	31730-64	5 g
			31730-22	25 g
Sodium *1*-Hexanesulfonate	C_6H_{13}-	SP	31529-24	5 g
			31529-82	25 g
Sodium *1*-Heptanesulfonate	C_7H_{15}-	SP	31528-34	5 g
			31528-92	25 g
Sodium *1*-Octanesulfonate	C_8H_{17}-	SP	31729-04	5 g
			31729-62	25 g
Sodium *1*-Nonanesulfonate	C_9H_{19}-	SP	31626-44	5 g
Sodium *1*-Decanesulfonate	$C_{10}H_{21}$-	SP	31429-34	5 g
Sodium *1*-Dodecanesulfonate	$C_{12}H_{25}$-	SP	31426-64	5 g
Sodium Lauryl Sulfate	**	SP	31623-32	25 g

0.5M Solution

Sodium *1*-Butanesulfonate	C_4H_9-	SP	31332-84	5×10 ml
Sodium *1*-Hexanesulfonate	C_6H_{13}-	SP	31532-64	10 ml
			31532-06	5×10 ml
Sodium *1*-Octanesulfonate	C_8H_{17}-	SP	31733-34	10 ml
			31733-76	5×10 ml

● for Acid Samples $(C_4H_9)_4N^+X^-$

Product Name	X-:	Grade	Product Number	PKG Size
Tetra-*n*-butylammonium Bromide	-Br	SP	32824-72	25 g
Tetra-*n*-butylammonium Chloride	-Cl	EP	32935-51	1 g
			32935-64	5 g
			32935-22	25 g
Tetra-*n*-butylammonium Hydrogensulfate	$-HSO_4$	GR	32924-62	25 g
Tetra-*n*-butylammonium Iodide	-I	SP	32905-54	5 g
			32905-12	25 g
Tetra-*n*-butylammonium Perchlorate	$-ClO_4$	SP	32906-44	5 g
			32906-02	25 g
Tetra-*n*-butylammonium Phosphate	$-H_2PO_4$	SP	32929-54	5 g

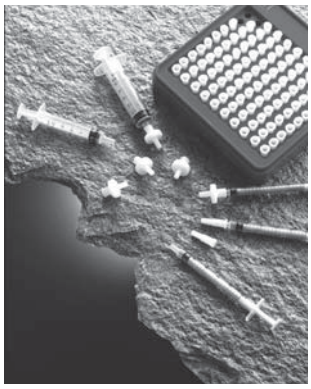
0.5M Solution

Tetra-*n*-butylammonium Phosphate	$-H_2PO_4$	SP	32926-26	10 ml
			32926-84	5x10 ml

2. Prefiltration Tool for Liquid Chromatography

Cosmonice Filter

- For sample filtration
- Just attach a filter on top of a syringe



W Series (aqueous solution)

W series are installed new material of low-adsorptive and low-extractive durapore-filter (poly vinylidenedifluoride, PVDF) which can be used for the various solvents. So they are able to minimize the loss of the proteins in the small amount of sample, and to prevent from secondary contamination at the prefiltration.

S Series (organic solvents)

S series are installed teflon-filter (poly tetrafluoroethylene, PTFE) shows strong resistance for solvents, acids, and alkalis. It is the best for the prefiltration of the sample extracted with solvents such as chloroform, tetrahydrofuran, and so on.

Please refer to Technical Information 4, Sample pretreatment for HPLC at page 193.

Ordering Information

● Cosmonice Filter

Product Name	Diameter (mm)	Pore Size (μm)	Process Volume	Hold-up Volume	Product Number	PKG Size
Cosmonice Filter W (aqueous)	4	0.45	1 ml or less	< 10 μl	06543-04	100 pkg
	13	0.45	0.5~10 ml	< 30 μl	06544-94	100 pkg
Cosmonice Filter S (solvent)	4	0.45	1ml or less	< 10 μl	06541-24	100 pkg
	13	0.45	0.5~10 ml	< 30 μl	06542-14	100 pkg

[Connection] Inlet: luer-lock, Outlet: luer-slip, Connectable needles
*housing : polyethylene

Cosmospin Filter

- For Sample filtration
- Easy to use by centrifugation
- Omunipore hydrophilic PTFE membrane filter

Please refer to Technical Information 4, Sample pretreatment for HPLC at page 193.

Ordering Information

● Cosmospin Filter

Product Name	Pore Size (μm)	Maximum Sample Volume	Hold-up Volume	Maximum Centrifugal Force	Rotor Size (fixed-angle)	Filtration Area	Color	Product Number	PKG Size
Cosmospin Filter G	0.2	0.4 ml	5 μl	5000 xg	1.5 ml	0.2 cm^2	Brown	06549-44	100 pkg
Cosmospin Filter H	0.45	0.4 ml	5 μl	5000 xg	1.5 ml	0.2 cm^2	White	06540-34	100 pkg

Dimention: Diameter 10.6 mm x Length 45 mm Membrane: Omnipore Hydrophilic PTFE Sample reservoir and collection tube: Polypropylene

Chemical Compatibility

Solvent	Cosmonice W series	Cosmonice S series	Cosmospin	Solvent	Cosmonice W series	Cosmonice S series	Cosmospin
Acetic acid, 98%	+	+	+	Hydrogen gas	+	+	+
Acetone	–	+	+	Hydrogen peroxide (3%)	+	+	
Acetonitrile	+	+	+	Hydraulic oil (5606)	+	+	+
Ammonia solution (6N)	+	+	+	Hypo (photo)	+	+	+
Ammounium hydroxide (conc.)	+	+	–	Isopropyl acetate	+	+	+
Amyl alcohol	+	+	+	Isopropyl alcohol	+	+	+
Benzene	+	+	–	Kerosene	+	+	+
Benzyl alcohol	+	+	–	Methanol	+	+	+
Boric acid	+		+	Methyl ethyl ketone	–	+	+
Butyl acetate		+		Methyl isobutyl ketone	+	+	–
Carbon tetrachloride	+	+	+	*2*-Methyl-*1*-propanol	+	+	+
Chloroform	+	+	+	Nitric acid (6N)	+	+	
Cyclohexanone	–	+	–	Nitrobenzene	+	+	–
Dichloromethane	+	+	–	Ozone gas	–	+	–
Dimethylacetamide	–	+	+	Paraldehyde		+	
Dimethylformamide	+	+	+	Pentane	+	+	–
Dimethylsulfoxide	–	+	–	Petroleum ether	+	+	
Dioxane	+	+	+	Phenol (water saturation)	+	+	–
DMSO	–	+	–	Phosphate buffer solution	+		+
Ethers	+	+	+	*2*-Propanol	+	+	+
Ethyl acetate	+	+	+	Pyridine	–	+	+
Ethyl alcohol	+	+	+	Seawater	+	+	+
Ethyl cello solve	+	+	+	Silicone oils	+	+	+
Ethylene glycol	+	+	+	Sodium hydroxide (conc.)	+	+	+
Formamide	+	+	+	Sulfuric acid (6N)		+	
Freon, TF or PCA solvent	+	+	+	Toluene	+	+	–
Gasoline	+	+	+	THF	–	+	–
Glycerine (Glycerol)	+	+	+	Trichloroacetic acid	+	+	+
Helium gas		+	+	Trichloroethane	+	+	–
Hexane	+	+	–	Trichloroethylene	+	+	–
Hydrochloride (6N)	+	+	+	TFA	+	+	–
Hydrofluoric acid	–	+	–	Xylene	+	+	+

+ : Recommended, – : Not recomended, (blank) : Not data available

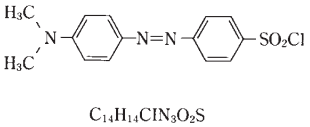
Labeling Reagents

Introduction

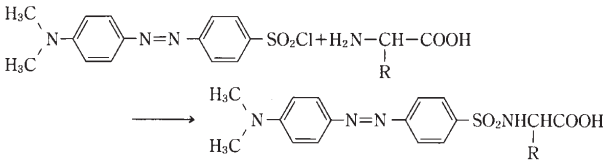
Labeling reagents enable high sensitivity detection and improved separation of amino acids or lipids. These reagents are available with various types.

● Dabsyl Chloride (*4*-Dimethylaminoazobenzene-*4'*-sulfonyl Chloride) (Visible labeling reagent)

Dabsyl Chloride is a dark-red crystalline, amino group labeling reagent with melting point of is 185-188°C. It is insoluble in water and slightly soluble in acetone. Dabsyl derivatives with strong absorption in the visible part (maximum absorption of 430 nm) are formed when the chemical bonding occurs between Dabsyl Chloride and amines, or amino acids. It shows 100-fold increased sensitivity comparing to DNBC (*3,5*-Dinitrobenzoyl Chloride), and the same sensitivity as with fluorescence labeling reagent, Dansyl Chloride.

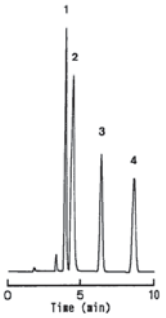


$C_{14}H_{14}ClN_3O_2S$
M.W.323.80



COSMOSIL Application Data

Column:	$5C_{18}$-MS-II
Column size:	4.6mmI.D.-150mm
Mobile phase:	Methanol/ 20mmol/l Phosphoric Acid = 70/30
Flow rate:	1.0 ml/min
Temperature:	30°C
Detection:	UV430nm, 2.56AUFS
Sample:	1; Dabsyl-Glycine 2; Dabsyl-*L*-Alanine 3; Dabsyl-*L*-Valine 4; Dabsyl-*L*-Leucine



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E.g.,) 1) Labeling of amino acids

Dissolve 60 nmol amino acid in 0.3 ml of sodium hydrogen carbonate buffer (pH 8.9, 0.1 mol/l). Add 0.3 ml of 10 umol/l acetone solution of Dabsyl Chloride Agitate the solution at 70°C for 6 minutes.

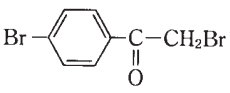
2) Labeling of Peptides

Dissolve 300 nmol sample in 0.3 ml of sodium hydrogen carbonate buffer (pH 8.9). Add 0.5 ml of 10 µmol/l acetone solution of Dabsyl Chloride. Agitate the solution at 70°C for 6 minutes. Vacuum dry the solution. Add 1.5 ml of 5.7 *N*-hydrochloric acid, and hydrolyze it at 105°C for 3 hours.

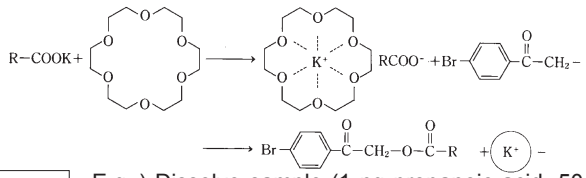
Ref. 1) J.K.Lin,J.Y.Chang,*Anal.Chem.*,47,1634 (1975)
2) J.Y.Chang,E.H.Creaser,*J.Chromatogr.*,116,215 (1976)
3) J.K.Lin,C.C.Lai,*Anal.Chem.*,52,630 (1980)

● *p*-Bromophenacyl Bromide (PBPB) (UV labeling reagent)

PBPB is a white crystalline, carboxylic acid labeling reagent soluble in alcohol. The melting point is 108-111°C. Using crown ether as the catalyst, fatty acid is reacted with PBPB to form p-bromophenacyl ester. The shorter the alkyl chain length of fatty acids is, the higher sensitivity can be achieved. (E.g., 1 ng propanoic acid, 50 ng stearic acid)

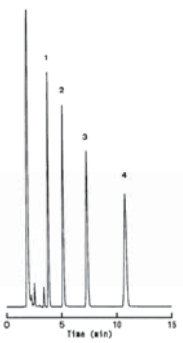


$C_8H_6Br_2O$
M.W.227.94



COSMOSIL Application Data

Column:	$5C_{18}$-MS-II
Column size:	4.6mmI.D.-150mm
Mobile phase:	Acetonitrile
Flow rate:	1.0 ml/min
Temperature:	30°C
Detection:	UV254nm, 2.56AUFS
Sample:	1; Lauric Acid p-Bromophenacyl Ester 2; Myristic Acid p-Bromophenacyl Ester 3; Palmitic Acid p-Bromophenacyl Ester 4; Stearic Acid p-Bromophenacyl Ester



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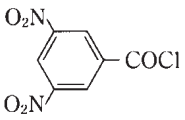
E.g.,) Dissolve sample (1 ng propanoic acid, 50 ng stearic acid) as potassium salt in 10 ml acetonitrile. Add mix solution of PBPB and *18*-crown-*6* (20:1) in the sample solution, and incubate at 80°C for 15 minutes. Cool down the reaction solution to room temperature.

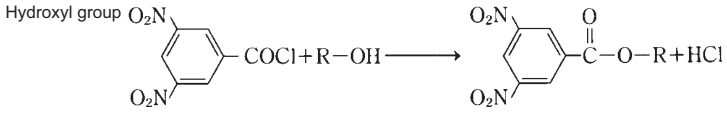
Ref. 1) Durst,H.D.,Milano,M.,Kikta,E.J.,Cinnelly,S.A., Grushka,E., *Anal Chem.*,47,1797 (1975)
2) F.A.Fitzpatrick,*Anal.Chem.*,48,499 (1976)
3) P.C.Bossle,J.J.Martin,E.W.Sarver,H.Z.Sommer,*J.Chromatogr.*, 267,209 (1983)
4) Kihara,S. Rokushika,H. Hatano,*Bunseki Kagaku*,33,647 (1984)
5) S.Konosu,A.Shinagawa,K.Yamaguchi,*Bulletin of Japanese Society of Scientific Fisheries*,52,869 (1986)

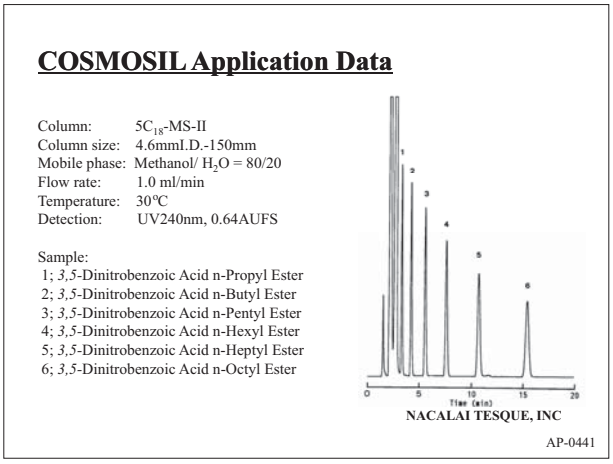
● *3,5*-Dinitrobenzoyl Chloride (DNBC) (UV labeling reagent)

DNBC is a yellow crystalline labeling reagent soluble in alcohol, ether and benzene. The melting point is 67-69°C. DNBC reacts with hydroxyl group (R-OH) to produce *3,5*-dinitrobenzoyl ether. It also reacts with amino group (R-NH2, R-NH-R') to produce *3,5*-dinitrobenzamide.



$C_7H_3ClN_2O_5$
M.W.230.56





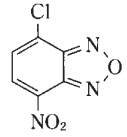
E.g.,) Dissolve a few mg of a sample in 5ml tetrahydrofuran. Add 40 mg of DNBC and 2-3 drops of pyridine, and seal to react at 60°C for 1 hour. Cool down the reaction solution to room temperature. If pyridine or triethylamine is used to eliminate hydrochloric acid, clean up is required before analysis.

How to clean up: Eliminate solvent and extract ether. Wash ether layer with dilute hydrochloric acid and water.

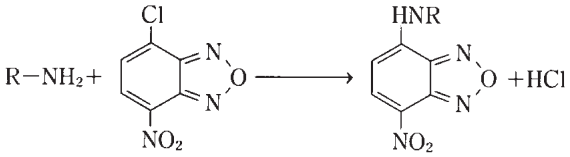
Ref. 1) T.H.Jupile,*Am.Lab.*,8,85 (1976)
2) M.A.Carey,H.E.Persinger,*J.Chromatogr.Sci.*,10,573 (1972)
3) Y. Suzuki, N. Tsuchiya, *Bunseki Kagaku*,30,240 (1981)
4) L.J.Elrod,L.B.White,S.G.Spanton,D.G.Stroz,P.J.Cugier, *L.A.Luka,Anal.Chem*,56,1786 (1984)

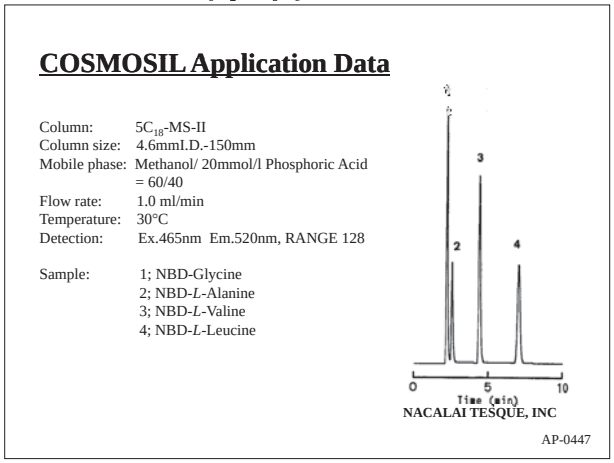
● NBD Chloride (*7*-Chloro-*4*-nitrobenz-*2*-oxa-*1,3*-diazole) (Fluorescence-labeling reagent)

NBD Chloride is a yellow crystallin labeling reagent soluble in alcohol. It is more soluble in water than dansyl chloride. The melting point is 95-98°C. NBD Chloride reacts with amino group ($R-NH_2$, R-NH-R'). It is also reacts with mercapto group (R-SH).



$C_6H_2ClN_3O_3$



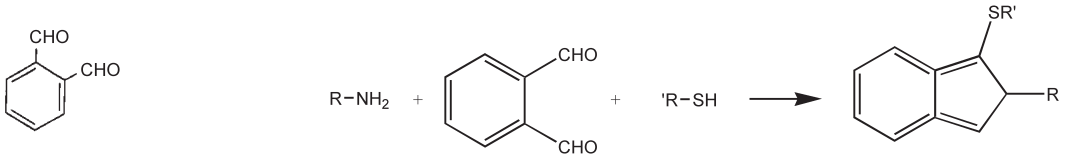


E.g.,) Prepare 25-500 ul of methanol solution containing 1-20 ug of amine. Add 0.05% methanol solution containing 4-8 times higher molar concentration of NBD Chloride than the concentration of amine. Add 50-100 ul of 0.1M sodium hydrogen carbonate, and incubate at 55°C for 1-5 hours. Cool down the reaction solution to room temperature.

Ref. 1) J.F.Lawrenoe,R.W.Frei,*Anal.Chem.*,44,2046 (1972)
2) H.F,Van,A.Heyndrickx,*Anal.Chem.*,46,286 (1974)
3) H,J.Klimisch,L,Stadlen,*J.Chromatogr.*,90,141 (1974)
4) J.H.Wolfram,*J.Chromatogr.*,132,37 (1977)
5) Y.Nishikawa,K.Kuwata,*Anal,Chem.*,57,1864 (1985)

● *o*-Phthalaldehyde,recryst (OPA) (Fluorescence-labeling reagent)

OPA is a yellow crystallin labeling reagent soluble in water and most of organic solvents and insoluble in ether. The melting point is 56–57°C. OPA reacts with primary amine and thiol compound.



COSMOSIL Application Data

Column: $5C_{18}$-MS-II
Column size: 4.6mmI.D.-150mm
Mobile phase: Methanol/ 20mmol/l Phosphoric Acid = 70/30
Flow rate: 1.0 ml/min
Temperature: 30°C
Detection: Ex.340nm Em.450nm, RANGE 128

Sample: 1; OPA-Glycine
2; OPA-*L*-Alanine
3; OPA-*L*-Valine
4; OPA-*L*-Leucine

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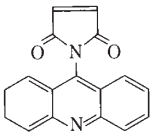
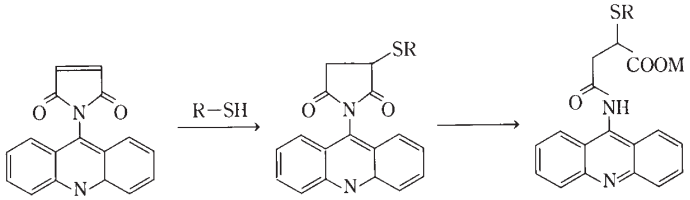
E.g.,) Prepare 0.05ml of 10% hydrochloric acid containing 5 ml of sample as acidic solution. Add 2 ml of 4 mg/ml methanol solution of OPA. Add 0.05 ml of *2*-mercaptoethanol and 3 ml of pH9 carbonate buffer. Incubate at room temperature for one minute and filter.

● N-(*9*-Acridinyl)maleimide (NAM) (Fluorescence-labeling reagent)

NAM is is a yellow crystallin labeling reagent. The melting point is 257–262°C. NAM react with mercapto group.



$C_{17}H_{10}N_2O_2$
M.W.274.28



E.g.,) Prepare 2 ml of 1 mmol sample solution containing 0.4ml of 30% sodium hydroxide and 1 ml of 0.2 mol boric acid buffer (pH8.8). Add 0.5 ml of 10 mmol acetone solution of NAM. Incubate at room temperature for 30 minutes.

Ref. 1) Y.Nara,K.Tujimura,*Agrlc.Biol.Chem.*,42,793 (1978)
2) H.Takahashi,Y.Nara,K.Tujimura,*Agric.Biol.Chem.*,43,1493 (1979)
3) H.Takahashi,T. Yoshida,Meguro,*Bunsekikagaku*,30,339 (1981)

Ordering Information

Product name	Grade	Storage	Product number	PKG Size
Dabsyl Chloride	SP	Room temp.	10427-91	1 g
p-Bromophenacyl Bromide (PBPB)	GR	Refrigerator	05802-92	25 g
3,5-Dinitrobenzoyl Chloride (DNBC)	SP	Dark and Cool	13530-44	5 g
NBD Chloride	SP	Refrigerator	24113-61	1 g
o-Phthalaldehyde (OPA)	SP	Refrigerator	27824-61	1 g
			27824-74	5 g
			27824-32	25 g
N-(*9*-Acridinyl) maleimide (NAM)	SP	Refrigerator	00842-64	50 mg

3. Column Care Products

Introduction

It is important to preserve a column by washing it with suitable cleaning methods before storing it under appropriate conditions to obtain stable data and prolong the column life time.

Applicable column

Cleaning Solution Kit and Storage Solution for Reversed Phase HPLC Columns are only applicable to reversed phase HPLC columns such as COSMOSIL 5C$_{18}$-MS-II, AR-II, PAQ, Cholester, πNAP, PYE and PBB-R. Please note that these products are not applicable for Sugar-D, HILIC, Normal phase and Ion Exchange columns.

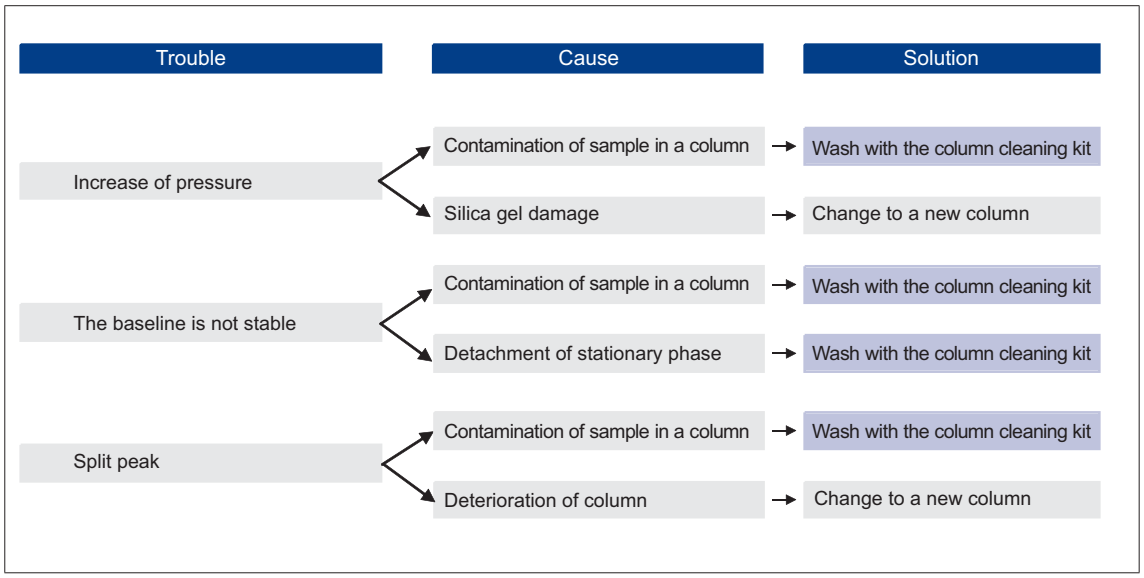
Cleaning Solution Kit for Reversed Phase HPLC Columns

Components

Product Name	Main Compositions	PKG Size	Quantity	Container
Cleaning Solution A	Methanol	500 ml	2	Brown Glass Bottle
Cleaning Solution B	Tetrahydrofuran, Methanol	500 ml	1	Brown Glass Bottle

Application

Cleaning Solution Kit for Reversed Phase HPLC Columns is designed for washing away contaminant adsorption and stationary phase shedding. If you experience the following symptoms, please try their corresponding solution first.



Procedure

(For 4.6 mm I.D. x 150 mm)

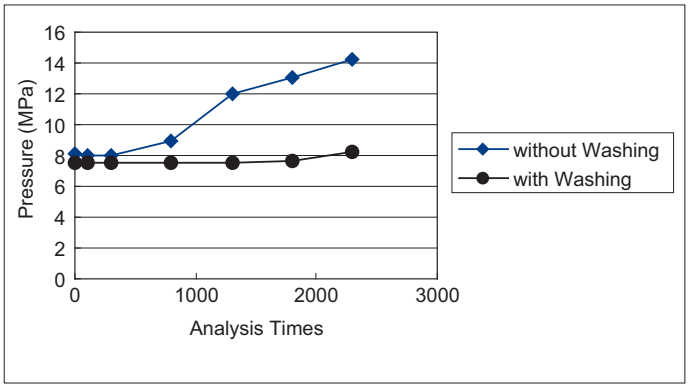
(1) Replace solvent with HPLC-grade distilled water (1 ml/min, 30 min).
(*This step is for mobile phase containing high concentration buffer. If you are using a salt-free mobile phase, please start from step (2).)

(2) Run the “Cleaning Solution A” through the column for 15 min at a flow rate of 1ml/min.

(3) Run the “Cleaning Solution B” through the column at a flow rate of 1ml/min until the baseline becomes stable (approx. 15 min).

(4) Run the “Cleaning Solution A” through the column for 15 min. The column is ready for storage.

Example of pressure difference between washed and unwashed columns



The figure shows pressure comparison between washed and unwashed columns using Cleaning Solution Kit. Repeated analysis of natural products was conducted using COSMOSIL 5C$_{18}$-MS-II 4.6 mm I.D. x 150 mm.

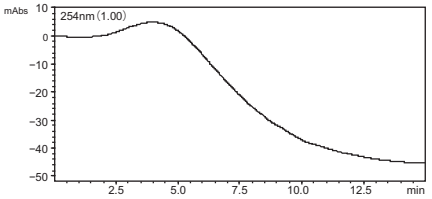
(Condition)
Column: COSMOSIL 5C$_{18}$-MS-II（4.6 mm I.D. × 150 mm）
Mobile phase: Methanol / H_2O = 70 / 30, Flow rate: 1.0ml/min, Temp.: 40°C

As shown in the figure above, the column pressure increases if you use column continuously without washing. If you wash the column, you can extend the column life time and ease the pressure burden on your HPLC equipment.

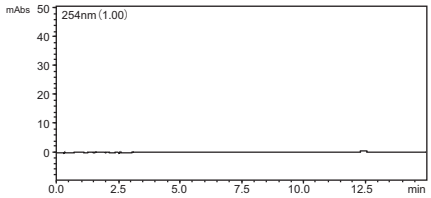
Example of a Stable Baseline

The baseline may be unstable if sample components with very long retention remain in the column or the stationary pha-se shedding occurs. Especially when analyzing crude compounds that have components with wide range of chemcal characteristics, some unwanted components may be strongly retained in the column and slowly elute out in subsequent runs. The resulting unstable baseline can be eliminated by washing the column with the Cleaning Solution Kit.

Before Washing



After Washing



Ordering Information

Product Name	Grade	Product No..	PKG Size
Cleaning Solution for Reversed Phase HPLC Columns	SP	08966-30	1 kit

Storage Solution for Reversed Phase HPLC Columns

Application

Storage Solution for Reversed Phase HPLC Columns is designed for storing column under suitable condition.

Procedure

(For 4.6 mm I.D. x 150 mm)

(1) Replace solvent with HPLC-grade distilled water. (1 ml/min, 30 min)

(*This step is for mobile phase containing high concentration buffer. If you are using a salt-free mobile phase, please start from step(2).)

(2) Run the “Storage Solution" through the column for 15 min at a flow rate of 1ml/min, and store.

Ordering Information

Product Name	Grade	Product No	PKG Size
Storage Solution for Reversed Phase HPLC Columns	SP	08967-20	1 kit (500 ml)

4. COSMOSIL HPLC Accessories

Ordering Information

COSMOSIL Guard Cartridge Holder

Product number	PKG Size
38009-79	1 PKG

Guard Cartridge Holder is required for Guard Cartridge.

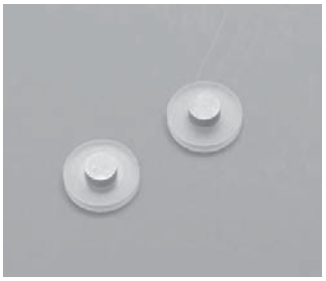
COSMOSIL Column Prefilter

Product number	PKG Size
39361-19	1 PKG

COSMOSIL Column Prefilter employs filter with smaller pore size (1 µm) than that of column frit (2 µm) .

COSMOSIL Column Spare Filter for Prefilter

Product number	PKG Size
39539-09	2 PKG



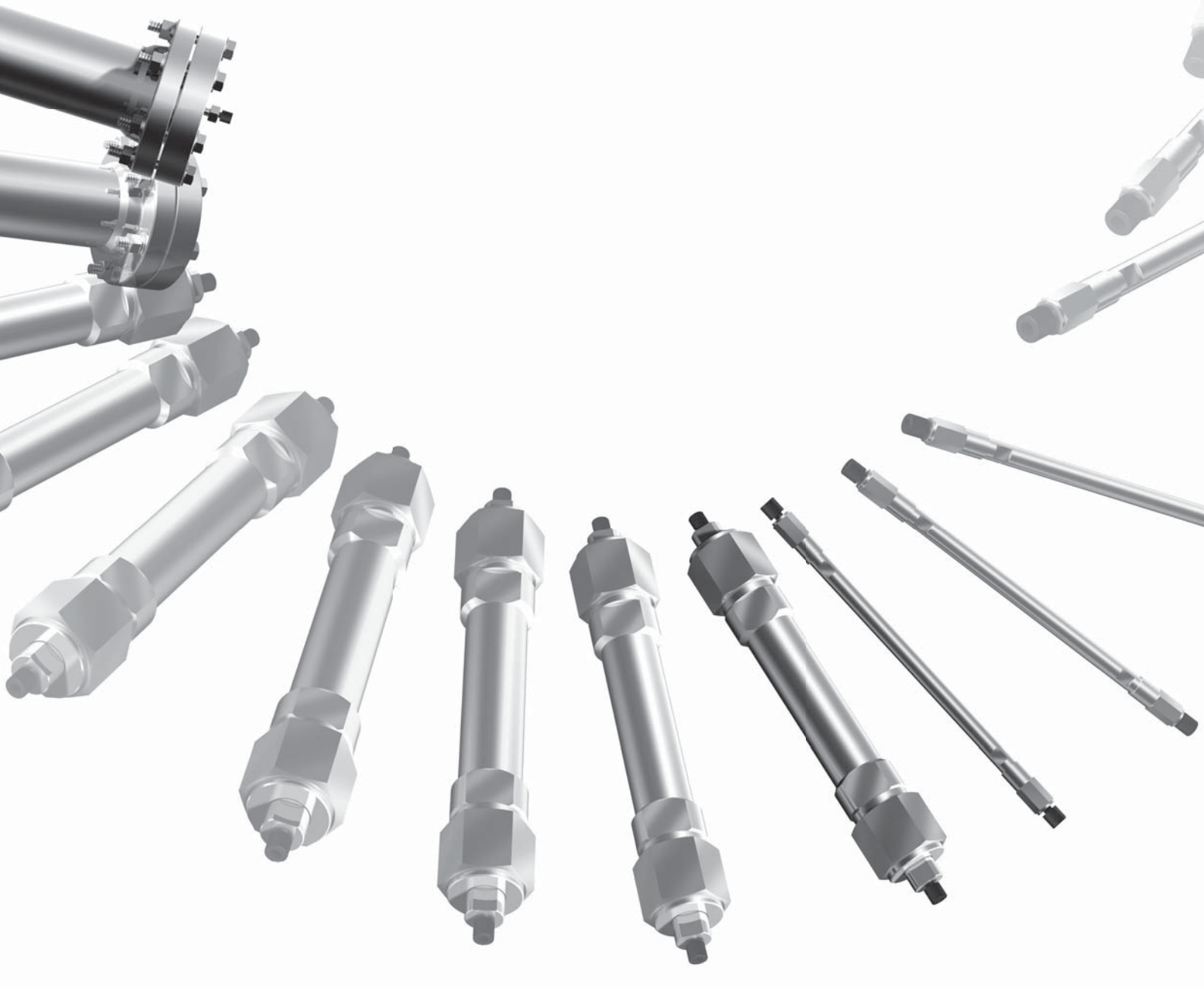
Column spare filter for prefilter

COSMOSIL Column Connecting Tube

Product number	PKG Size
37843-69	1 PKG

for connecting columns

V Applications



1. COSMOSIL Applications

Introduction

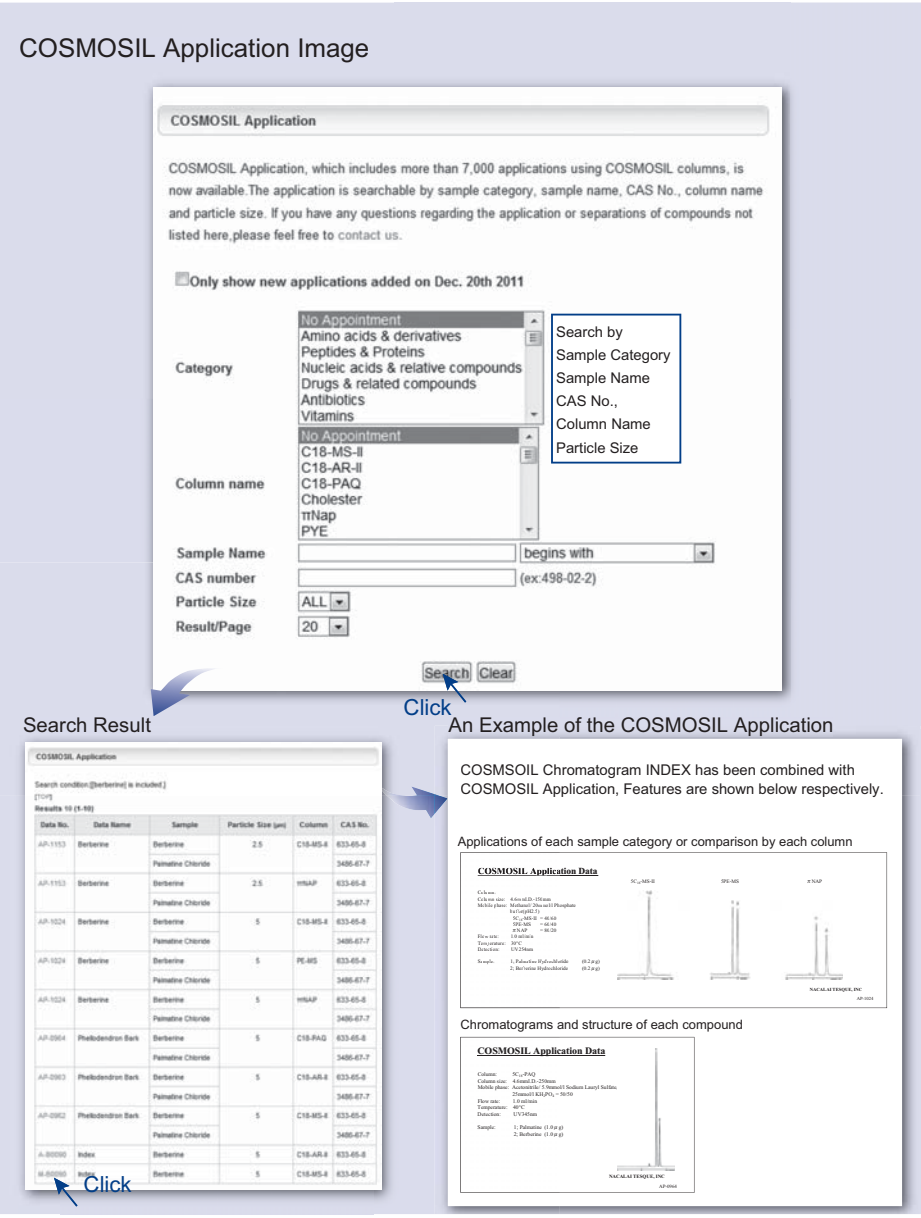
COSMOSIL Application has more than 7,000 applications using COSMOSIL columns. Setting optimal HPLC experimental parameters is the one of the most important processes that requires experience and time. COSMOSIL Application provides you with sample analysis conditions with widely used ODS columns and other specialty columns. For more information, please visit COSMSOIL Application page on our website at http://www.nacalai.co.jp/global/cosmosil/.

- Over 7,000 applications
- Easy to search

How to Search

The application is searchable by sample category, sample name, CAS No., column name and particle size. If you have any questions regarding the application or separations of compounds not listed here, please feel free to contact us at info.intl@nacalai.com

COSMOSIL Application Image

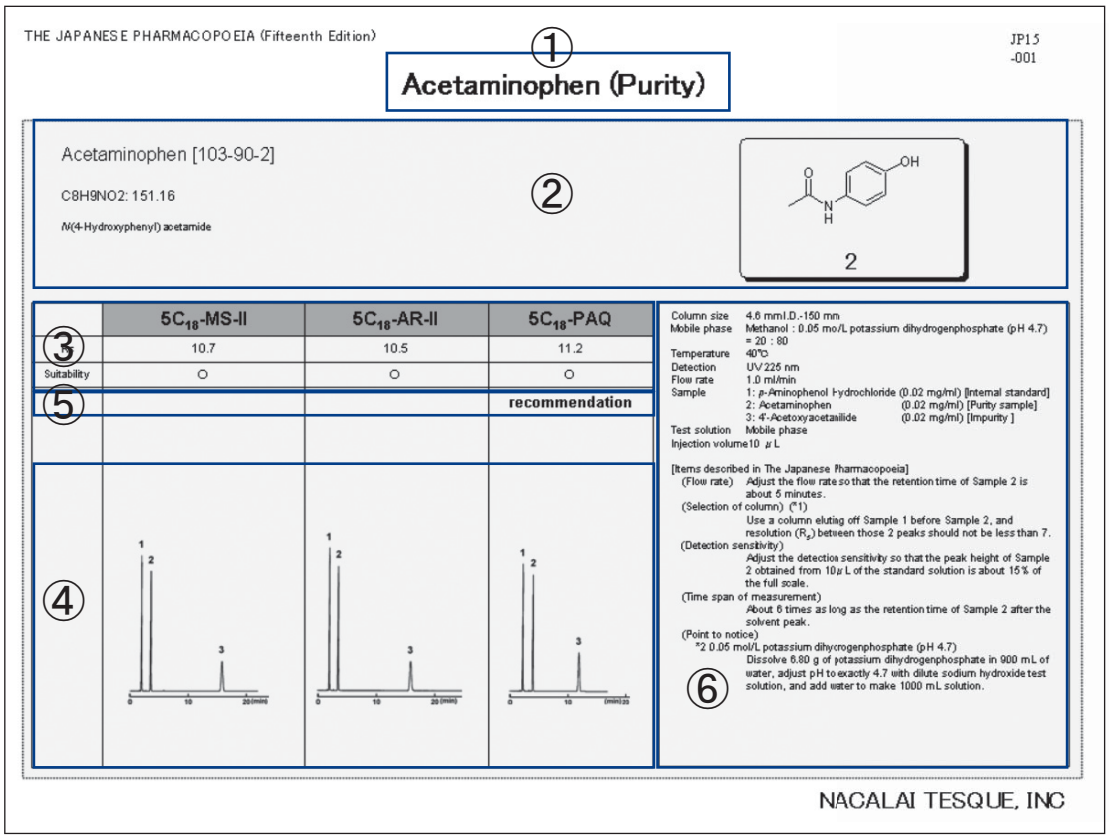


2. Applications of Drug Substances in the Japanese Pharmacopeia, 15th

Introduction

We have prepared drug analysis HPLC data using three different COSMOSIL C_{18} columns as specified in Japanese Pharmacopoeia, 15th version. 5 µm silica gel column size 4.6mm I.D. x 150 mm or 250 mm were used with reagent grade samples. Furthermore, we analyzed reference drug substances with internal standard and impurities. The data are available at our web site at http://www.nacalai.co.jp/global/cosmosil/, or search “COSMOSIL Japanese Pharmacopoeia” on the web. An example of the application is shown after page 92.

Interpretation of Application



① Substance Name

② Substance Information

③ Suitability (○ : suitable, × : unsuitable, ** : depend on condition)

④ HPLC Chromatogram

⑤ Recommended Column

Recommended column is determined by resolution of internal standard substances or impurities, theoretical plate number, peak symmetry of the target sample and analysis conditions.

⑥ Condition and Japanese Pharmacopeia Description

3. COSMOSIL Applications

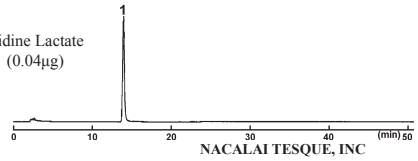
(1) Drugs

● Acrinol

COSMOSIL Application Data

Column: $5C_{18}$-PAQ
Column size: 4.6mmI.D.-250mm
Mobile phase: Acetonitrile/ 4.6mmol/l Sodium *1*-Octanesulfonate, 65mmol/l NaH_2PO_4 (pH2.8 with H_3PO_4) =30/70
Flow rate: 1.0 ml/min
Temperature: 25°C
Detection: UV268nm

Sample:
1; *6,9*-Diamino-*2*-ethoxyacridine Lactate 〔Acrinol〕 (0.04µg)



NACALAI TESQUE, INC

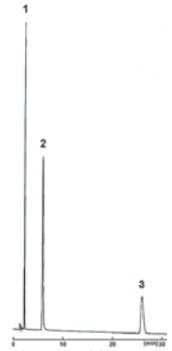
AP-0457

● Azathioprine

COSMOSIL Application Data

Column: $5C_{18}$-AR-II
Column size: 4.6mmI.D.-150mm
Mobile phase: Methanol/ 25mmol/l KH_2PO_4 (pH2.5 with H_3PO_4) = 20/80
Flow rate: 1.0 ml/min
Temperature: 40°C
Detection: UV296nm

Sample:
1; *6*-Mercaptopurine (0.16µg)
2; Azathioprine (0.16µg)
3; Benzoic Acid (9.6µg)



NACALAI TESQUE, INC

AP-0471

● Aztreonam

COSMOSIL Application Data

Column: $5C_{18}$-AR-II
Column size: 4.6mmI.D.-250mm
Mobile phase: Methanol/ 5mmol/l Tetra-*n*-butylammonium Hydrogensulfate(pH3.0 with 0.5mol/l Na_2HPO_4) = 35/65
Flow rate: 1.0 ml/min
Temperature: 40°C
Detection: UV280nm

Sample:
1; *p*-Aminobenzoic Acid (0.5µg)
2; Aztreonam (5.0µg)

NACALAI TESQUE, INC

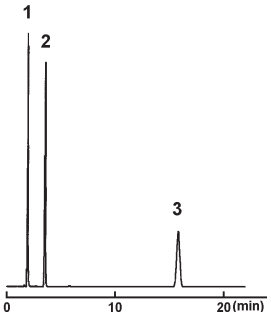
AP-0474

● Acetaminophen

COSMOSIL Application Data

Column: $5C_{18}$-MS-II
Column size: 4.6mmI.D.-150mm
Mobile phase: Methanol/ 50mmol/l KH_2PO_4 (pH4.7 with NaOH) = 20/80
Flow rate: 1.0 ml/min
Temperature: 40°C
Detection: UV225nm

Sample:
1; *p*-Aminophenol (0.2µg)
2; *p*-Acetamidophenol (0.2µg)
3; *4'*-Acetoxyacetanilide (0.2µg)



NACALAI TESQUE, INC

AP-0452

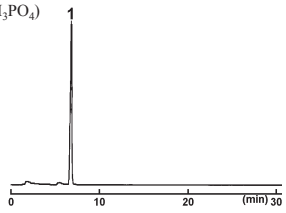
(1) Drugs

● Atenolol

COSMOSIL Application Data

Column: $5C_{18}$-AR-II
Column size: 4.6mmI.D.-150mm
Mobile phase: Methanol/Tetrahydrofuran/
4.6mmol/l Sodium *1*-Octanesulfonate,
1.2mmol/l Tetra-*n*-butylammonium Hydrogensulfate,
25mmol/l KH_2PO_4(pH3.0 with H_3PO_4)
= 9/1/40
Flow rate: 1.0 ml/min
Temperature: 25°C
Detection: UV226nm

Sample: 1; Atenolol (0.1µg)



NACALAI TESQUE, INC

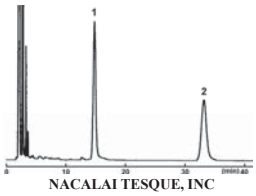
AP-0468

● Amikacin Sulfate

COSMOSIL Application Data

Column: $5C_{18}$-PAQ
Column size: 4.6mmI.D.-250mm
Mobile phase: Methanol/ 20mmol/l KH_2PO_4
(pH6.5 with KOH) = 72/28
Flow rate: 1.5 ml/min
Temperature: 35°C
Detection: UV340nm

Sample:
1; Amikacin Sulfate *2,4,6*-trinitrobenzenesufonic Acid Derivative (0.6µg)
2; Kanamycin Sulfate *2,4,6*-trinitrobenzenesufonic Acid Derivative (0.6µg)



NACALAI TESQUE, INC

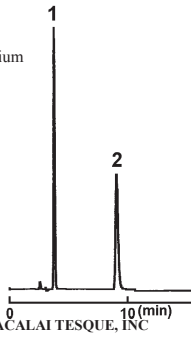
AP-0460

● Meglumine Sodium Amidotrizoate

COSMOSIL Application Data

Column: $5C_{18}$-MS-II
Column size: 4.6mmI.D.-250mm
Mobile phase: Acetonitrile/ 6.25mmol/l Tetra-*n*-butylammonium Phosphate, 50mmol/l K_2HPO_4
(pH7.0 with H_3PO_4) = 20/80
Flow rate: 1.0 ml/min
Temperature: 25°C
Detection: UV254nm

Sample: 1; Diatrizoic Acid (0.25µg)
2; Acetrizoic Acid (0.30µg)



NACALAI TESQUE, INC

AP-0739

● Amlodipine Besilate

COSMOSIL Application Data

Column: $5C_{18}$-MS-II
Column size: 4.6mmI.D.-150mm
Mobile phase: Methanol/ 30mmol/l KH_2PO_4 = 65/35
Flow rate: 0.5 ml/min
Temperature: 30°C
Detection: UV237nm

Sample: 1; Amlodipine (0.56µg)
2; Isobutyl *p*-Hydroxybenzoate (0.60µg)

NACALAI TESQUE, INC

AP-1084

● Amoxicillin

COSMOSIL Application Data

Column: $5C_{18}$-PAQ
Column size: 4.6mmI.D.-150mm
Mobile phase: Methanol/ 10mmol/l CH_3COONa
(pH4.5 with CH_3COOH) = 5/95
Flow rate: 1.0 ml/min
Temperature: 30°C
Detection: UV230nm

Sample: 1; Amoxicillin (3.0µg)

NACALAI TESQUE, INC

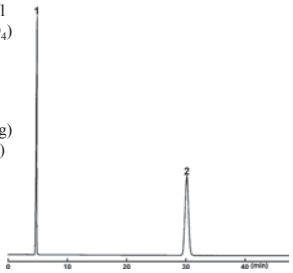
AP-0463

● Ampicillin

COSMOSIL Application Data

Column: $5C_{18}$-MS-II
Column size: 4.6mmI.D.-150mm
Mobile phase: 10% Acetonitrile/ 90% 50mmol/l $(NH_4)_2HPO_4$ (pH5.0 with H_3PO_4)
Flow rate: 1.0 ml/min
Temperature: 25°C
Detection: UV230nm

Sample: 1; Ampicillin (10µg)
2; Guaiacol Glycerol Ether (5µg)



NACALAI TESQUE, INC

AP-0464

(1) Drugs

● Isoxsuprine Hydrochloride

COSMOSIL Application Data

Column: $5C_{18}$-MS-II
Column size: 4.6mmI.D.-150mm
Mobile phase: Methanol/ 32.6mmol/l $(NH_4)_2HPO_4$, 18.4mmol/l Sodium *1*‐Pentanesulfonate (pH2.5 with H_3PO_4) = 40/60
Flow rate: 1.0 ml/min
Temperature: 40°C
Detection: UV269nm

Sample: 1; Isoxsuprine (2.0µg)

NACALAI TESQUE, INC

AP-1103

● Isoniazid

COSMOSIL Application Data

Column: $5C_{18}$-MS-II
Column size: 4.6mmI.D.-250mm
Mobile phase: 10mmol/l-Sodium *1*-Tridecanesulfonate-Methanol/ 50mmol/l Phosphate buffer (pH2.5) =60/40
Flow rate: 0.5 ml/min
Temperature: 40°C
Detection: UV265nm

Sample: 1; Isonicotinic Acid [Pyridine-*4*-carboxylic Acid] (0.4µg)
2; Isonicotinic Hydrazide [Isoniazid] (0.5µg)

NACALAI TESQUE, INC

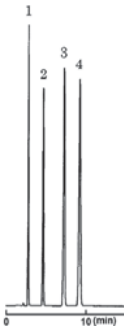
AP-0702

● Idoxuridine

COSMOSIL Application Data

Column: $5C_{18}$-PAQ
Column size: 4.6mmI.D.-150mm
Mobile phase: Methanol/ H_2O = 13/87
Flow rate: 1.0 ml/min
Temperature: 30°C
Detection: UV254nm

Sample: 1; *2'*-Deoxyuridine (0.3µg)
2; *5*-Iodouracil (1.2µg)
3; *5*-Iododeoxyuridine [Idoxuridine] (3.0µg)
4; Sulfathiazole (0.5µg)



NACALAI TESQUE, INC

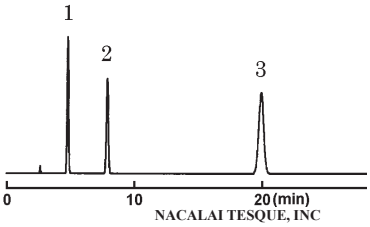
AP-0694

● Idoxuridine

COSMOSIL Application Data

Column: $5C_{18}$-PAQ
Column size: 4.6mmI.D.-150mm
Mobile phase: Methanol/ H_2O = 4/96
Flow rate: 1.0 ml/min
Temperature: 30°C
Detection: UV254nm

Sample:
1; *2'*-Deoxyuridine (0.04µg)
2; *5*-Iodouracil (0.12µg)
3; *5*-Iododeoxyuridine [Idoxuridine] (0.40µg)



NACALAI TESQUE, INC

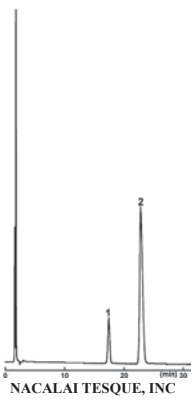
AP-0697

● Ipratropium Bromides

COSMOSIL Application Data

Column: $5C_8$-MS
Column size: 4.6mmI.D.-150mm
Mobile phase: Acetonitrile/ 0.5%H_3PO_4/ Methanesulfonic Acid = 120/1000/10
Flow rate: 1.0 ml/min
Temperature: 30°C
Detection: UV210nm

Sample: 1; Ipratropium Bromide Derivative
2; Ipratropium Bromide



NACALAI TESQUE, INC

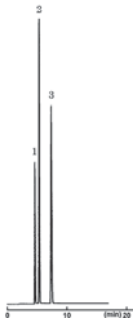
AP-0701

● Indomethacin

COSMOSIL Application Data

Column: $5C_{18}$-MS-II
Column size: 4.6mmI.D.-250mm
Mobile phase: Methanol/ 0.1%H_3PO_4 = 80/20
Flow rate: 1.0 ml/min
Temperature: 25°C
Detection: UV254nm

Sample: 1; *4*-Chlorobenzoic Acid (1.0µg)
2; Butyl *p*-Hydroxybenzoate (0.6µg)
3; Indometacin(Indomethacin) (1.0µg)



NACALAI TESQUE, INC

AP-0698

(1) Drugs

● Estradiol Benzoate

COSMOSIL Application Data

Column: $5C_{18}$-MS-II
Column size: 4.6mmI.D.-150mm
Mobile phase: Acetonitrile/ H_2O = 70/30
Flow rate: 1.0 ml/min
Temperature: 35°C
Detection: UV230nm

Sample: 1; Progesterone (0.15µg)
2; Estradiol Benzoate (0.65µg)

NACALAI TESQUE, INC

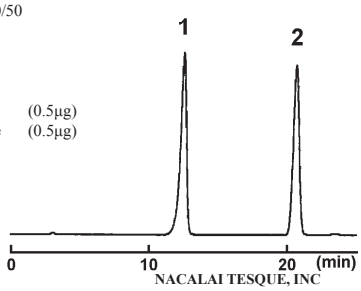
AP-0647

● Estriol

COSMOSIL Application Data

Column: $5C_{18}$-MS-II
Column size: 4.6mmI.D.-150mm
Mobile phase: Methanol/ H_2O = 50/50
Flow rate: 0.5 ml/min
Temperature: 25°C
Detection: UV280nm

Sample: 1; Estriol (0.5µg)
2; Methyl Benzoate (0.5µg)



NACALAI TESQUE, INC

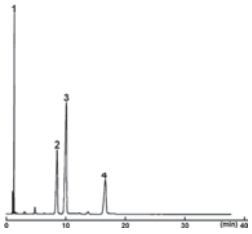
AP-0644

● Epirubicin Hydrochloride

COSMOSIL Application Data

Column: 5TMS-MS
Column size: 4.6mmI.D.-250mm
Mobile phase: 6.9mmol/l Sodium Lauryl Sulfate-H_2O/Acetonitrile/Methanol/H_3PO_4 = 540/290/170/1
Flow rate: 2.0 ml/min
Temperature: 35°C
Detection: UV254nm

Sample:
1; 2-Naphthalenesulfonic Acid (5.0µg)
2; Doxorubicin (5.0µg)
3; Epirubicin (10µg)
4; Daunorubicin (5.0µg)



NACALAI TESQUE, INC

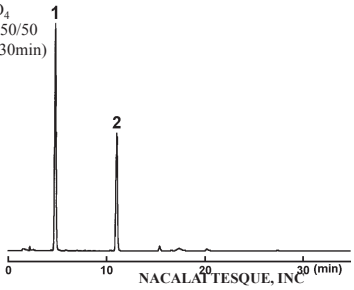
AP-0643

● Oxytocin

COSMOSIL Application Data

Column: $5C_{18}$-MS-II
Column size: 4.6mmI.D.-150mm
Mobile phase: A; 100mmol/l NaH_2PO_4
B; Acetonitrile/H_2O = 50/50
B conc. 30→60%(0→30min)
Flow rate: 1.0 ml/min
Temperature: 25°C
Detection: UV220nm

Sample:
1; [Arg^8]-Vasopressin (5.0µg)
2; Oxytocin Acetate (5.0µg)



NACALAI TESQUE, INC

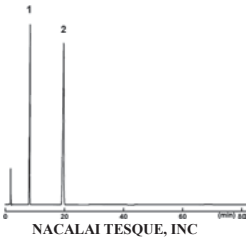
AP-0793

● Omeprazole

COSMOSIL Application Data

Column: $5C_8$-MS
Column size: 4.6mmI.D.-150mm
Mobile phase: Acetonitrile/ 7.9mmol/l Na_2HPO_4, 1.35mmol/l NaH_2PO_4(pH7.6 with 1% H_3PO_4) = 11/29
Flow rate: 1.0 ml/min
Temperature: 30°C
Detection: UV280nm

Sample: 1; Omeprazole (0.10µg)
2; *1,2*-Dinitrobenzene (0.25µg)



NACALAI TESQUE, INC

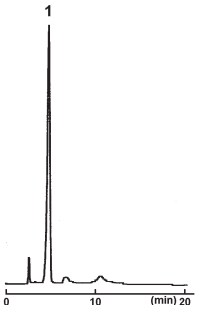
AP-1121

● Captopril

COSMOSIL Application Data

Column: $5C_{18}$-MS-II
Column size: 4.6mmI.D.-250mm
Mobile phase: Metahnol/ 0.1%Acetic Acid = 50/50
Flow rate: 1.0 ml/min
Temperature: 25°C
Detection: UV220nm

Sample: 1; Captopril (2.6µg)



NACALAI TESQUE, INC

AP-0530

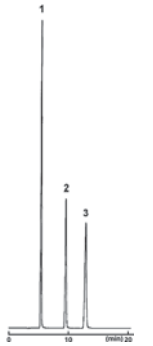
(1) Drugs

● Gabexate Mesilate

COSMOSIL Application Data

Column: $5C_{18}$-MS-II
Column size: 4.6mmI.D.-150mm
Mobile phase: Methanol/ (0.1%Sodium Lauryl Sulfate/ 0.5%Sodium *1*-Heptanesulfonate/ Acetic Acid = 200/20/1) = 71/29
Flow rate: 0.5 ml/min
Temperature: 25°C
Detection: UV245nm

Sample:
1; Ethyl *p*-Hydroxybenzoate (0.39µg)
2; Butyl *p*-Hydroxybenzoate (0.39µg)
3; Gabexate Mesilate (0.75µg)



NACALAI TESQUE, INC

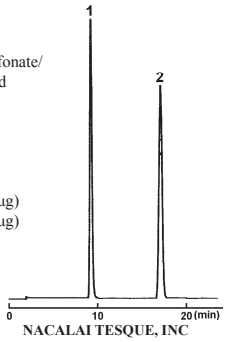
AP-0673

● Camostat Mesilate

COSMOSIL Application Data

Column: $5C_{18}$-AR-II
Column size: 4.6mmI.D.-150mm
Mobile phase: Metahnol/ (0.2%Sodium *1*-Heptane Sulfonate/ 0.1%Sodium Lauryl Sulfate/ Acetic Acid = 1000/500/10) = 55/45
Flow rate: 1.0 ml/min
Temperature: 25°C
Detection: UV265nm

Sample:
1; Camostat (1.02µg)
2; Butyl *p*-Hydroxybenzoate (0.54µg)



NACALAI TESQUE, INC

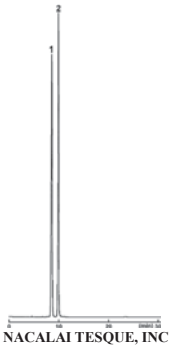
AP-0528

● Carbazochrome Sodium Sulfonate

COSMOSIL Application Data

Column: $5C_{18}$-PAQ
Column size: 4.6mmI.D.-250mm
Mobile phase: Ethanol/ 10.4mmol/l $(NH_4)_2HPO_4$ = 75/925(pH7 with H_3PO_4)
Flow rate: 1.0 ml/min
Temperature: 40°C
Detection: UV360nm

Sample:
1; Carbazochrome Sodium Sulfonate (1.0µg)
2; Carbazochrome (1.0µg)



NACALAI TESQUE, INC

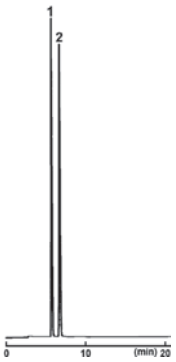
AP-0535

● Carbidopa

COSMOSIL Application Data

Column: $5C_{18}$-PAQ
Column size: 4.6mmI.D.-250mm
Mobile phase: Ethanol/ 50mmol/l NaH_2PO_4 = 5/95 (pH2.7 with H_3PO_4)
Flow rate: 1.0 ml/min
Temperature: 30°C
Detection: UV280nm

Sample:
1; (-)-*3*-(*3,4*-Dihydroxyphenyl)-*2*-Methyl-*L*-Alanine [*L*-α-Methyl DOPA] (10µg)
2; *S*(-)-Carbidopa (10µg)



NACALAI TESQUE, INC

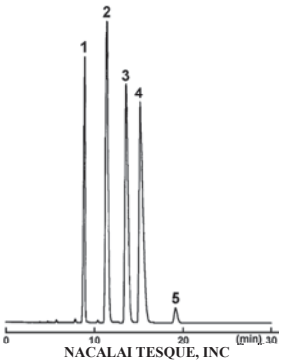
AP-0538

● Quinidine Sulfate and Quinine Hydrochloride

COSMOSIL Application Data

Column: $5C_{18}$-PAQ
Column size: 4.6mmI.D.-250mm
Mobile phase: Acetonitrile/ 0.088%Acetic Acid, 0.16%Methanesulfonic Acid, 0.22%Diethylamine = 10/90
Flow rate: 1.0 ml/min
Temperature: 30°C
Detection: UV235nm

Sample:
1; Cinchonidine (1.0µg)
2; Quinidine (10µg)
3; Quinine (10µg)
4; Hydroquinidine (10µg)
5; Dihydroquinine



NACALAI TESQUE, INC

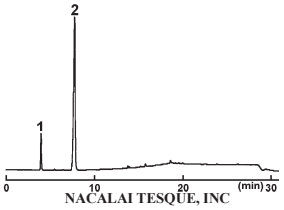
AP-0828

● Potassium Clavulanate

COSMOSIL Application Data

Column: $5C_{18}$-PAQ
Column size: 4.6mmI.D.-150mm
Mobile phase: A; 50mmol/l NaH_2PO_4(pH4.0 with H_3PO_4)
B; Methanol/ 50mmol/l NaH_2PO_4 (pH4.0 with H_3PO_4) = 50/50
B conc. 0%(0-4 min), 0→100%(4→15 min), 100%(15-25min)
Flow rate: 1.0 ml/min
Temperature: 40°C
Detection: UV230nm

Sample:
1; Clavulanic Acid
2; Amoxicillin



NACALAI TESQUE, INC

AP-0810

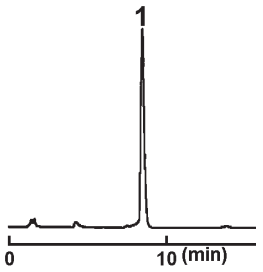
(1) Drugs

● Clarithromycin

COSMOSIL Application Data

Column: $5C_{18}$-MS-II
Column size: 4.6mmI.D.-150mm
Mobile phase: Acetonitrile/ 67mmol/l KH_2PO_4 = 35/65
Flow rate: 1.0 ml/min
Temperature: 50°C
Detection: UV210nm

Sample: 1; Clarithromycin (2.8µg)



NACALAI TESQUE, INC

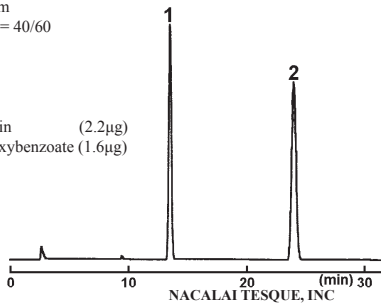
AP-0578

● Griseofulvin

COSMOSIL Application Data

Column: $5C_{18}$-MS-II
Column size: 4.6mmI.D.-250mm
Mobile phase: Acetonitrile/ H_2O = 40/60
Flow rate: 1.0 ml/min
Temperature: 25°C
Detection: UV254nm

Sample: 1; (+)-Griseofulvin (2.2µg)
2; Butyl *p*-Hydroxybenzoate (1.6µg)



NACALAI TESQUE, INC

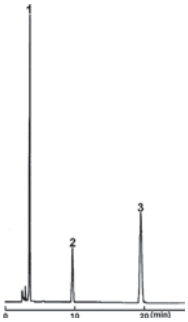
AP-0676

● Clindamycin Phosphate

COSMOSIL Application Data

Column: $5C_8$-MS
Column size: 4.6mmI.D.-250mm
Mobile phase: Acetonitrile/ 100mmol/l KH_2PO_4 (pH2.5 with H_3PO_4) = 22/78
Flow rate: 1.0 ml/min
Temperature: 25°C
Detection: UV210nm

Sample: 1; Lincomycin (4.0µg)
2; Clindamycin Phosphate (4.0µg)
3; Methyl *p*-Hydroxybenzoate (0.3µg)



NACALAI TESQUE, INC

AP-0584

● Clindamycin Hydrochloride

COSMOSIL Application Data

Column: $5C_{18}$-MS-II
Column size: 4.6mmI.D.-150mm
Mobile phase: Acetonitrile/ 50mmol/l KH_2PO_4 (pH7.5 with KOH) = 45/55
Flow rate: 1.0 ml/min
Temperature: 40°C
Detection: UV210nm

Sample: 1; Clindamycin (15µg)

NACALAI TESQUE, INC

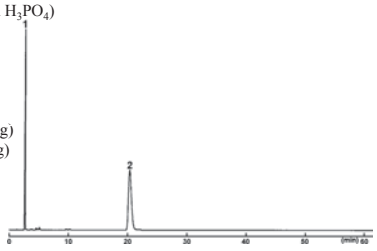
AP-0581

● Cloxacillin Sodium

COSMOSIL Application Data

Column: $5C_{18}$-MS-II
Column size: 4.6mmI.D.-150mm
Mobile phase: Acetonitrile/ 50mmol/l $(NH_4)_2HPO_4$ = 25/75(pH4.0 with H_3PO_4)
Flow rate: 1.5 ml/min
Temperature: 25°C
Detection: UV230nm

Sample:
1; Guaiacol Glycerol Ether (5.0µg)
2; Cloxacillin (10µg)



NACALAI TESQUE, INC

AP-0585

● Clofibrate

COSMOSIL Application Data

Column: 5CN-MS
Column size: 4.6mmI.D.-250mm
Mobile phase: Hexane/2-Propanol/Acetic Acid = 1970/30/1
Flow rate: 1.0 ml/min
Temperature: 30°C
Detection: UV275nm

Sample: 1; Clofibrate (2.0µg)
2; *p*-Chlorophenol (0.12µg)
3; *p*-Ethoxyphenol (0.12µg)

NACALAI TESQUE, INC

AP-0588

(1) Drugs

● Clofibrate

COSMOSIL Application Data

Column: $5C_{18}$-AR-II
Column size: 4.6mmI.D.-250mm
Mobile phase: Acetonitrile/ 0.1%Acetic Acid = 60/40
Flow rate: 1.5 ml/min
Temperature: 25°C
Detection: UV275nm

Sample:
1; *4*-Chlorophenol (4.0µg)
2; Ibuprofen (60µg)
3; Clofibrate (10µg)

NACALAI TESQUE, INC

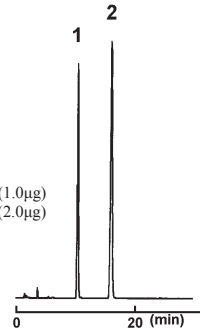
AP-0590

● Clobetasol Propionate

COSMOSIL Application Data

Column: $5C_{18}$-MS-II
Column size: 4.6mmI.D.-150mm
Mobile phase: Acetonitrile/ Methanol/ 50mmol/l NaH_2PO_4 (pH2.5 with H_3PO_4) = 47.5/10/42.5
Flow rate: 1.0 ml/min
Temperature: 30°C
Detection: UV240nm

Sample:
1; Clobetasol Propionate (1.0µg)
2; Beclomethasone Dipropionate (2.0µg)



NACALAI TESQUE, INC

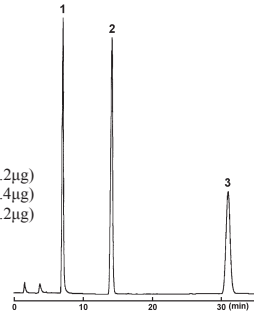
AP-1098

● Cloperastine Hydrochloride

COSMOSIL Application Data

Column: $5C_{18}$-AR-II
Column size: 4.6mmI.D.-150mm
Mobile phase: Methanol/ 100mmol/l KH_2PO_4, 0.16%Perchloric Acid = 60/40
Flow rate: 1.0 ml/min
Temperature: 25°C
Detection: UV222nm

Sample:
1; Cloperastine (0.2µg)
2; Benzophenone (0.4µg)
3; *4*-Chlorobenzophenone (0.2µg)



NACALAI TESQUE, INC

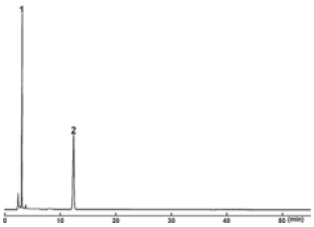
AP-0593

● Chlorpheniramine Maleate

COSMOSIL Application Data

Column: $5C_{18}$-PAQ
Column size: 4.6mmI.D.-250mm
Mobile phase: Acetonitrile/ 74.5mmol/l $(NH_4)H_2PO_4$, 14.7mmol/l H_3PO_4 = 20/80
Flow rate: 1.0 ml/min
Temperature: 25°C
Detection: UV225nm

Sample:
1; Maleic Acid (Impurity)
2; Chlorpheniramine (0.06µg)



NACALAI TESQUE, INC

AP-0568

● Chlorpropamide

COSMOSIL Application Data

Column: $5C_{18}$-AR-II
Column size: 4.6mmI.D.-250mm
Mobile phase: Acetonitrile/ 1%Acetic Acid = 50/50
Flow rate: 1.0 ml/min
Temperature: 25°C
Detection: UV240nm

Sample:
1; *1*-(*p*-Chlorophenylsulfonyl)-*3*-Propylurea [Chlorpropamide] (1.0µg)

NACALAI TESQUE, INC

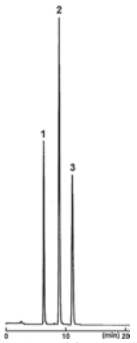
AP-0570

● Chlormadinone Acetate

COSMOSIL Application Data

Column: $5C_{18}$-MS-II
Column size: 4.6mmI.D.-150mm
Mobile phase: Acetonitrile/H_2O = 65/35
Flow rate: 0.5 ml/min
Temperature: 30°C
Detection: UV236nm

Sample:
1; Butyl *p*-Hydroxybenzoate (0.2µg)
2; *17α*-Hydroxyprogesterone *17*-Acetate (0.2µg)
3; Chlormadinone Acetate (0.8µg)



NACALAI TESQUE, INC

AP-0563

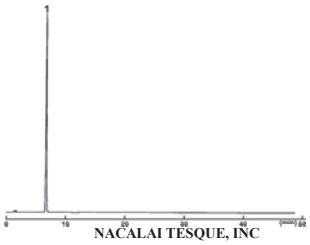
(1) Drugs

● Ketoprofen

COSMOSIL Application Data

Column: $5C_{18}$-AR-II
Column size: 4.6mmI.D.-150mm
Mobile phase: Acetonitrile/H_2O/ 0.5mol KH_2PO_4 (pH3.5 with H_3PO_4) = 43/55/2
Flow rate: 1.0 ml/min
Temperature: 25°C
Detection: UV233nm

Sample: 1; Ketoprofen (0.4µg)



NACALAI TESQUE, INC

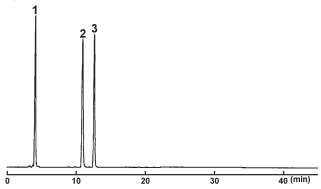
AP-0709

● Cortisone

COSMOSIL Application Data

Column: $5C_{18}$-MS-II
Column size: 4.6mmI.D.-150mm
Mobile phase: A; Acetonitrile/ H_2O =30/70
B; Acetonitrile/ H_2O =70/30
B conc. 10%(0-5min)
10→90%(5→25min)
90%(25-30min)
Flow rate: 1.0 ml/min
Temperature: 30°C
Detection: UV254nm

Sample:
1; Cortisone (3.6µg)
2; Cortisone *21*-Acetate (3.75µg)
3; Hydrocortisone Acetate (3.6µg)



NACALAI TESQUE, INC

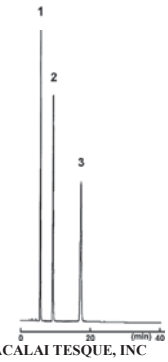
AP-0595

● Salicylic Acid

COSMOSIL Application Data

Column: $5C_{18}$-AR-II
Column size: 4.6mmI.D.-150mm
Mobile phase: Methanol/ 1.67% Acetic Acid = 40/60
Flow rate: 0.5 ml/min
Temperature: 35°C
Detection: UV270nm

Sample: 1; *p*-Hydroxybenzoic Acid (0.025µg)
2; Phenol (0.10µg)
3; Salicylic Acid (0.50µg)



NACALAI TESQUE, INC

AP-1123

● Santonin

COSMOSIL Application Data

Column: $5C_{18}$-MS-II
Column size: 4.6mmI.D.-150mm
Mobile phase: Methanol/H_2O = 50/50
Flow rate: 1.0 ml/min
Temperature: 25°C
Detection: UV254nm

Sample: 1; α-Santonin (0.50µg)
2; Ethyl *p*-Hydroxybenzoate (0.24µg)

NACALAI TESQUE, INC

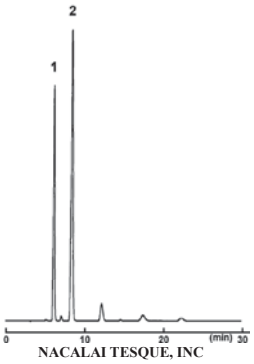
AP-0849

● Cyanocobalamins

COSMOSIL Application Data

Column: $5C_8$-MS
Column size: 4.6mmI.D.-250mm
Mobile phase: Methanol/ 70mmol/l Na_2HPO_4 (pH3.5 with H_3PO_4) = 53/147
Flow rate: 1.0 ml/min
Temperature: 30°C
Detection: UV361nm

Sample: 1; Vitamin B_{12} [Cyanocobalamin]
2; Cyanocobalamin Derivative



NACALAI TESQUE, INC

AP-1101

● Digitoxin

COSMOSIL Application Data

Column: $5C_{18}$-PAQ
Column size: 4.6mmI.D.-150mm
Mobile phase: Methanol/ H_2O = 75/25
Flow rate: 1.0 ml/min
Temperature: 30°C
Detection: UV230nm

Sample: 1; Digoxin (1.0µg)
2; Gitoxin (0.5µg)
3; Digitoxin (0.5µg)
4; Acenaphthene (0.03µg)

NACALAI TESQUE, INC

AP-0620

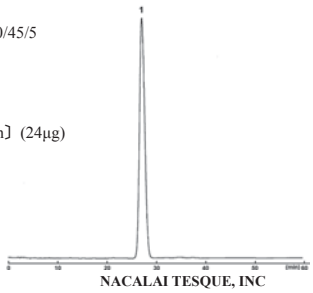
(1) Drugs

● Cyclosporin

COSMOSIL Application Data

Column: $5C_{18}$-PAQ
Column size: 4.6mmI.D.-250mm
Mobile phase: Acetonitrile/ 0.22%H_3PO_4 / *tert*-Butyl Methyl Ether = 50/45/5
Flow rate: 1.0 ml/min
Temperature: 50°C
Detection: UV210nm

Sample: 1; Ciclosporin〔Cyclosporin〕(24µg)



NACALAI TESQUE, INC

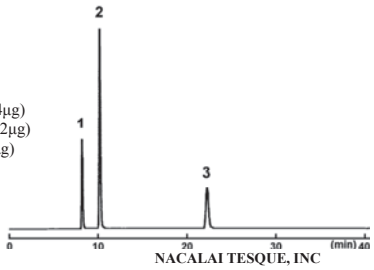
AP-0574

● Diclofenac Sodium

COSMOSIL Application Data

Column: $5C_{18}$-AR-II
Column size: 4.6mmI.D.-250mm
Mobile phase: Methanol/ 0.12%Acetic Acid = 70/30
Flow rate: 0.5 ml/min
Temperature: 40°C
Detection: UV240nm

Sample:
1; Ethyl *p*-Hydroxybenzoate (0.14µg)
2; *n*-Propyl *p*-Hydroxybenzoate (0.2µg)
3; Diclofenac (0.4µg)



NACALAI TESQUE, INC

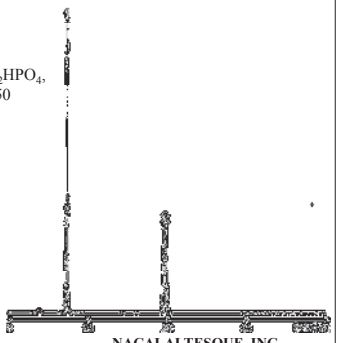
AP-0613

● Diclofenamide

COSMOSIL Application Data

Column: $5C_{18}$-MS-II
Column size: 4.6mmI.D.-250mm
Mobile phase: Acetonitrile/ 40mmol/l Na_2HPO_4, 52mmol/l NaH_2PO_4 = 50/50
Flow rate: 0.5 ml/min
Temperature: 30°C
Detection: UV280nm

Sample:
1; Dichlorophenamide (10µg)
2; Butyl *p*-Hydroxybenzoate (1.2µg)



NACALAI TESQUE, INC

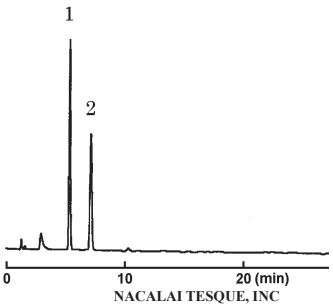
AP-0615

● Disulfiram

COSMOSIL Application Data

Column: $5C_{18}$-MS-II
Column size: 4.6mmI.D.-150mm
Mobile phase: Methanol/ H_2O = 70/30
Flow rate: 1.0 ml/min
Temperature: 25°C
Detection: UV210nm

Sample:
1; Benzophenone (0.05µg)
2; Tetraethylthiuram Disulfide [Disulfiram] (0.05µg)



NACALAI TESQUE, INC

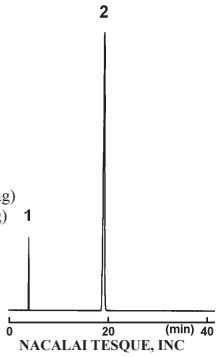
AP-0625

● Zidovudine

COSMOSIL Application Data

Column: $5C_{18}$-MS-II
Column size: 4.6mmI.D.-250mm
Mobile phase: Methanol/ H_2O = 20/80
Flow rate: 1.0 ml/min
Temperature: 30°C
Detection: UV265nm

Sample:
1; Thymine (0.08µg)
2; *3'*-Azido-*3'*-Deoxythymidine [Zidovudine] (2.0µg)



NACALAI TESQUE, INC

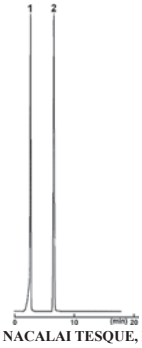
AP-1130

● Dipyridamole

COSMOSIL Application Data

Column: $5C_8$-MS
Column size: 4.6mmI.D.-150mm
Mobile phase: Methanol/ 7.5mmol/l KH_2PO_4 = 80/20
Flow rate: 1.0 ml/min
Temperature: 40°C
Detection: UV280nm

Sample: 1; Dipyridamole (2.8µg)
2; *p*-Terphenyl (1.2µg)



NACALAI TESQUE, INC

AP-0624

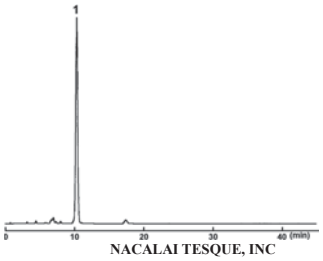
(1) Drugs

● Josamycin

COSMOSIL Application Data

Column: $5C_{18}$-AR-II
Column size: 4.6mmI.D.-150mm
Mobile phase: Acetonitrile/ 0.8mol Sodium Perchlorate (pH2.5 with HCl) = 40/60
Flow rate: 2.0 ml/min
Temperature: 40°C
Detection: UV231nm

Sample: 1; Josamycin (10µg)



NACALAI TESQUE, INC

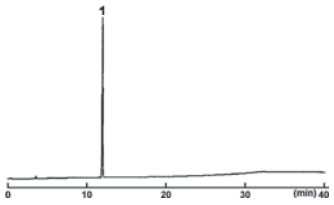
AP-0706

● Cilastatin Sodium Salt

COSMOSIL Application Data

Column: $5C_{18}$-AR-II
Column size: 4.6mmI.D.-250mm
Mobile phase: A; 0.1%H_3PO_4
B; Acetonitrile/ 0.1%H_3PO_4 = 30/70
B conc. 15→30%(0→30min), 100%(30-40min)
Flow rate: 2.0 ml/min
Temperature: 50°C
Detection: UV210nm

Sample: 1; Cilastatin (1.0µg)



NACALAI TESQUE, INC

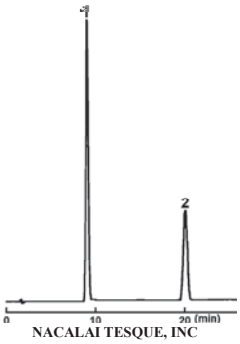
AP-0576

● Diltiazem Hydrochloride

COSMOSIL Application Data

Column: $5C_{18}$-MS-II
Column size: 4.6mmI.D.-150mm
Mobile phase: Methanol/Acetonitrile/ 118mmol/l CH_3COONa, 6.5mmol/l *d*-Camphorsulfonic Acid = 25/25/50(pH6.68)
Flow rate: 1.0 ml/min
Temperature: 50°C
Detection: UV240nm

Sample: 1; Diltiazem (3.0µg)
2; Phenyl Benzoate (2.0µg)



NACALAI TESQUE, INC

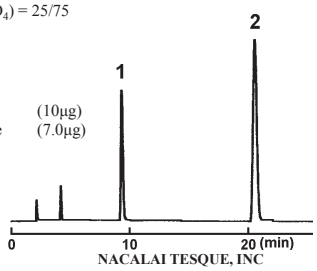
AP-0621

● Sulbactam Sodium

COSMOSIL Application Data

Column: $5C_{18}$-MS-II
Column size: 4.6mmI.D.-250mm
Mobile phase: Acetonitrile/ 5mmol/l Tetra *n*-Butylammonium Hydroxide(pH4.0 with H_3PO_4) = 25/75
Flow rate: 1.0 ml/min
Temperature: 35°C
Detection: UV220nm

Sample: 1; Sulbactam (10µg)
2; Ethyl *p*-Hydroxybenzoate (7.0µg)



NACALAI TESQUE, INC

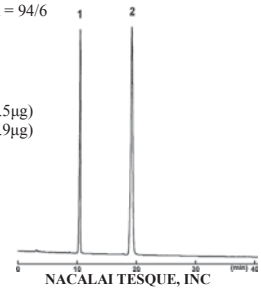
AP-0853

● Cetirizine Hydrochloride

COSMOSIL Application Data

Column: 5SL-II
Column size: 4.6mmI.D.-250mm
Mobile phase: Acetonitrile/ 40mmol/l Sulfuric Acid = 94/6
Flow rate: 1.0 ml/min
Temperature: 25°C
Detection: UV230nm

Sample: 1; Cetirizine (0.5µg)
2; *4*-Dimethylaminoantipyrine (0.9µg)



NACALAI TESQUE, INC

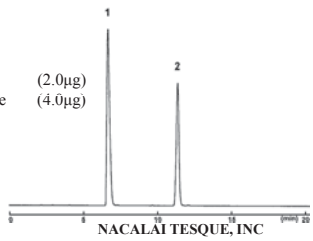
AP-1096

● Cetirizine Hydrochloride

COSMOSIL Application Data

Column: $5C_8$-MS
Column size: 4.6mmI.D.-250mm
Mobile phase: Acetonitrile/ 1.7mmol/l Sodium *1*-Heptanesulfonate = 42/58 (pH3.0 with 0.5mol/l Sulfuric Acid)
Flow rate: 1.0 ml/min
Temperature: 25°C
Detection: UV230nm

Sample: 1; Cetirizine (2.0µg)
2; Propyl *p*-Hydroxybenzoate (4.0µg)



NACALAI TESQUE, INC

AP-1097

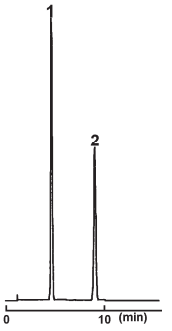
(1) Drugs

● Cefaclor

COSMOSIL Application Data

Column: $5C_{18}$-PAQ
Column size: 4.6mmI.D.-150mm
Mobile phase: Acetonitrile/ 50mmol/l KH_2PO_4 (pH3.4 with H_3PO_4) =6/94
Flow rate: 2.0 ml/min
Temperature: 25°C
Detection: UV254nm

Sample: 1; Cefaclor (2.0µg)
2; *p*-Aminoacetophenone (2.9µg)



NACALAI TESQUE, INC

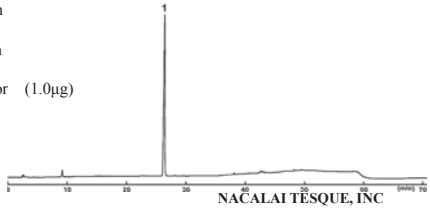
AP-0541

● Cefaclor

COSMOSIL Application Data

Column: $5C_{18}$-MS-II
Column size: 4.6mmI.D.-250mm
Mobile phase: A; 50mmol/l NaH_2PO_4(pH4.0 with H_3PO_4)
B; Acetonitrile/Buffer A = 45/55
B conc. 5→25%(0-30min), 25→100%(30-45min), 100%(45-55min)
Flow rate: 1.0 ml/min
Temperature: 25°C
Detection: UV220nm

Sample: 1; Cefaclor (1.0µg)



NACALAI TESQUE, INC

AP-0542

● Cefadroxil

COSMOSIL Application Data

Column: $5C_{18}$-MS-II
Column size: 4.6mmI.D.-250mm
Mobile phase: Methanol/ 10mmol/ KH_2PO_4 = 60/340
Flow rate: 1.0 ml/min
Temperature: 40°C
Detection: UV262nm

Sample: 1; Cefadroxil (1.3µg)

NACALAI TESQUE, INC

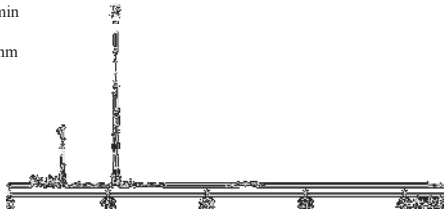
AP-0545

● Cephalothin Sodium Salt

COSMOSIL Application Data

Column: $5C_{18}$-MS-II
Column size: 4.6mmI.D.-250mm
Mobile phase: Acetonitrile/Ethanol/ 158mmol/l CH_3COONa, 0.076%Acetic Acid(pH5.9 with NaOH) =15/7/78
Flow rate: 1.0 ml/min
Temperature: 40°C
Detection: UV254nm

Sample:
1; Similar compound
2; Cephalothin Sodium (0.25µg)



NACALAI TESQUE, INC

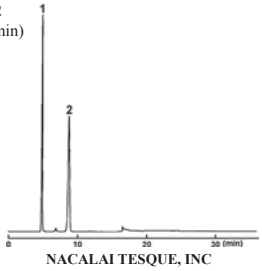
AP-0548

● Cefsulodin Sodium Salt

COSMOSIL Application Data

Column: $5C_{18}$-PAQ
Column size: 4.6mmI.D.-150mm
Mobile phase: A; Acetonitrile/1%$(NH_4)_2SO_4$ = 3/97
B; Acetonitrile/1%$(NH_4)_2SO_4$ = 8/92
B conc. 0%(0-14min), 100%(14-30min)
Flow rate: 1.0 ml/min
Temperature: 25°C
Detection: UV254nm

Sample: 1; Isonicotinamide (0.2µg)
2; Cefsulodin (0.2µg)



NACALAI TESQUE, INC

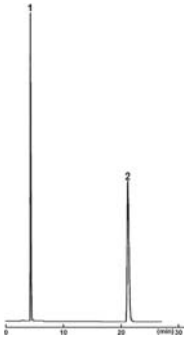
AP-0553

● Ceftazidime

COSMOSIL Application Data

Column: $5C_{18}$-AR-II
Column size: 4.6mmI.D.-250mm
Mobile phase: Acetonitrile/ 50mmol/l $(NH_4)H_2PO_4$ (pH3.5 with H_3PO_4) = 13/87
Flow rate: 1.0 ml/min
Temperature: 30°C
Detection: UV254nm

Sample: 1; Ceftazidime (2.5 µ g)
2; Acetanilide (2.5 µ g)



NACALAI TESQUE, INC

AP-0555

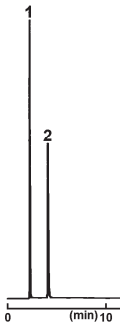
(1) Drugs

● Ceftazidime

COSMOSIL Application Data

Column: $5C_{18}$-MS-II
Column size: 4.6mmI.D.-250mm
Mobile phase: Acetonitrile/ 50mmol/l $(NH_4)H_2PO_4$ = 30/70(pH7.0 with NH_3)
Flow rate: 1.0 ml/min
Temperature: 40°C
Detection: UV254nm

Sample:
1; Ceftazidime (0.5µg)
2; Pyridine (0.5µg)



NACALAI TESQUE, INC

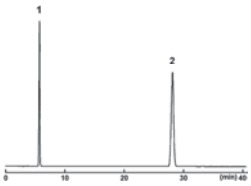
AP-0557

● Ceftriaxone Sodium

COSMOSIL Application Data

Column: $5C_{18}$-MS-II
Column size: 4.6mmI.D.-250mm
Mobile phase: 18.1mmol/l tetra-*n*-heptylammonium bromide-Acetonitrile / 4.1mmol/l Na_2HPO_4, 2.6mmol/l KH_2PO_4, 0.88mmol/l Citric Acid, 1.8mmol/l NaOH = 45/55
Flow rate: 1.0 ml/min
Temperature: 25°C
Detection: UV254nm

Sample:
1; Ceftriaxone (0.5µg)
2; Terephthalic Acid Diethyl Ester (0.9µg)



NACALAI TESQUE, INC

AP-0560

● Daunorubicin Hydrochloride

COSMOSIL Application Data

Column: $5C_{18}$-MS-II
Column size: 4.6mmI.D.-250mm
Mobile phase: Acetonitrile/ H_2O = 38/62 (pH2.2 with H_3PO_4)
Flow rate: 0.5 ml/min
Temperature: 25°C
Detection: UV254nm

Sample:
1; Daunorubicin (5.0µg)
2; *2*-Naphthalenesulfonic Acid (10µg)

NACALAI TESQUE, INC

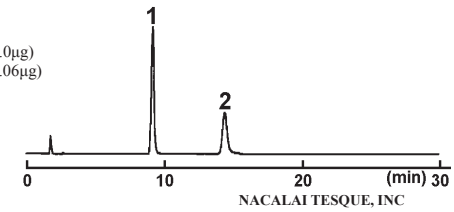
AP-0602

● Dantrolene Sodium

COSMOSIL Application Data

Column: 5SL-II
Column size: 4.6mmI.D.-250mm
Mobile phase: Hexane/Acetic Acid/Ethanol = 90/10/9
Flow rate: 2.0 ml/min
Temperature: 30°C
Detection: UV300nm

Sample:
1; Theophylline (1.0µg)
2; Dantrolene (0.06µg)



NACALAI TESQUE, INC

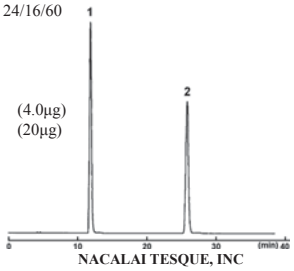
AP-0601

● Thiamine Hydrochloride

COSMOSIL Application Data

Column: $5C_{18}$-AR-II
Column size: 4.6mmI.D.-250mm
Mobile phase: Methanol/Acetonitrile/ 5mmol/l Sodium *1*-Octanesulfonate, 1%H_3PO_4 = 24/16/60
Flow rate: 1.0 ml/min
Temperature: 25°C
Detection: UV254nm

Sample:
1; Vitamin B_1 [Thiamine] (4.0µg)
2; Methyl Benzoate (20µg)



NACALAI TESQUE, INC

AP-0857

● Thiopental Sodiums

COSMOSIL Application Data

Column: $5C_{18}$-MS-II
Column size: 4.6mmI.D.-150mm
Mobile phase: Acetonitrile/ 7mmol/l KH_2PO_4 (pH3.0 with H_3PO_4) = 30/70
Flow rate: 1.5 ml/min
Temperature: 40°C
Detection: UV254nm

Sample:
1; Isopropyl *p*-Hydroxybenzoate (1.0µg)
2; *n*-Propyl *p*-Hydroxybenzoate (1.0µg)
3; Thiopental (Isomer)
4; Thiopental (4.8µg)

NACALAI TESQUE, INC

AP-0859

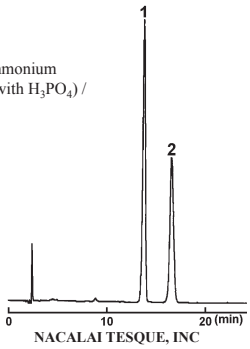
(1) Drugs

● Ticarcillin Sodium

COSMOSIL Application Data

Column: 5TMS-MS
Column size: 4.6mmI.D.-150mm
Mobile phase: Acetonitrile/ 5mmol/l Tetra-*n*-Butylammonium Bromide, 25mmol/l NaH_2PO_4(pH3.0 with H_3PO_4) / Acetic Acid = 225/1000/2.5
Flow rate: 1.0 ml/min
Temperature: 30°C
Detection: UV230nm

Sample: 1; *o*-Toluic Acid (2.5µg)
2; Ticarcillin (7.6µg)



NACALAI TESQUE, INC

AP-0862

● Tipepidine Hibenzate

COSMOSIL Application Data

Column: $5C_{18}$-MS-II
Column size: 4.6mmI.D.-150mm
Mobile phase: Tetrahydrofuran/ 1%Ammonium Acetate = 30/70
Flow rate: 1.0 ml/min
Temperature: 50°C
Detection: UV254nm

Sample: 1; Hibenzic Acid
2; Tipepidine
3; *n*-Propyl *p*-Hydroxybenzoate (0.6µg)

NACALAI TESQUE, INC

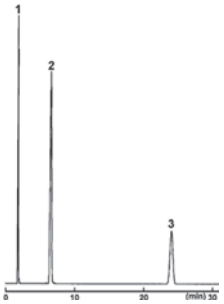
AP-0864

● Tipepidine Hibenzate

COSMOSIL Application Data

Column: $5C_{18}$-MS-II
Column size: 4.6mmI.D.-150mm
Mobile phase: Methanol/ 0.2%Ammonium Acetate = 65/35
Flow rate: 1.0 ml/min
Temperature: 40°C
Detection: UV254nm

Sample: 1; Hibenzic Acid
2; Tipepidine
3; Xanthene (0.8µg)



NACALAI TESQUE, INC

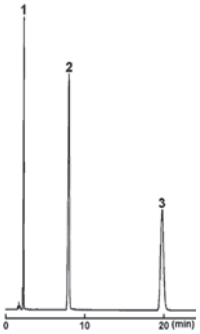
AP-0867

● Tipepidine Hibenzate

COSMOSIL Application Data

Column: $5C_{18}$-AR-II
Column size: 4.6mmI.D.-150mm
Mobile phase: (Acetonitrile/ *2*-Propanol = 2/1) / 0.1%Sodium Lauryl Sulfate, 0.2%H_3PO_4 = 50/50
Flow rate: 1.0 ml/min
Temperature: 40°C
Detection: UV254nm

Sample: 1; Hibenzic Acid
2; Tipepidine
3; Dibucaine (1.6µg)



NACALAI TESQUE, INC

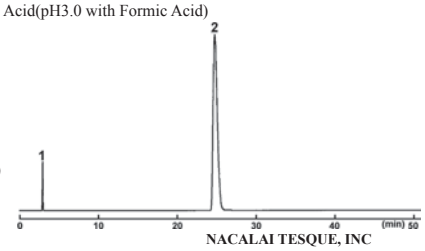
AP-0871

● Timolol Maleate

COSMOSIL Application Data

Column: 5PE-MS
Column size: 4.6mmI.D.-250mm
Mobile phase: Acetonitrile/Methanol/ 5mmol/l Sodium *1*-Hexanesulfonate, 0.3%Triethylamine, 0.4%Formic Acid(pH3.0 with Formic Acid) = 5/25/70
Flow rate: 1.0 ml/min
Temperature: 40°C
Detection: UV280nm

Sample:
1; Maleic Acid
2; Timolol (7.5µg)



NACALAI TESQUE, INC

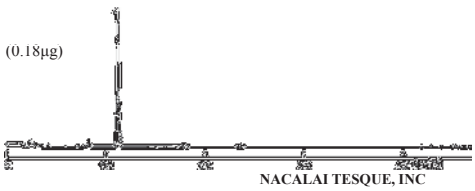
AP-0863

● Dexamethasone

COSMOSIL Application Data

Column: 5PE-MS
Column size: 4.6mmI.D.-250mm
Mobile phase: Acetonitrile/ 21mmol/l Ammonium Formate (pH3.6 with Formic Acid) = 33/67
Flow rate: 1.0 ml/min
Temperature: 30°C
Detection: UV254nm

Sample:
1; Dexamethasone (0.18µg)



NACALAI TESQUE, INC

AP-0611

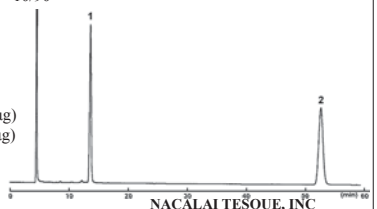
(1) Drugs

● Deferoxamine Mesilate

COSMOSIL Application Data

Column: $5C_{18}$-AR-II
Column size: 4.6mmI.D.-250mm
Mobile phase: *2*-Propanol/ 10.5mmol/l $(NH_4)_2HPO_4$, 1.05mmol/l EDTA, 5.6mmol/l Sodium *1*-Heptanesulfonate (pH2.8 with H_3PO_4) = 10/90
Flow rate: 0.5 ml/min
Temperature: 25°C
Detection: UV230nm

Sample:
1; Deferoxamine (6.4µg)
2; Methyl *p*-Hydroxybenzoate (1.6µg)



NACALAI TESQUE, INC

AP-0606

● Doxycycline Hydrochloride

COSMOSIL Application Data

Column: $5C_{18}$-MS-II
Column size: 4.6mmI.D.-250mm
Mobile phase: Methanol/ *N,N*-Dimethyloctylamine/ 100mmol/l NaH_2PO_4 = 550/3/450 (pH8.0 with NaOH)
Flow rate: 1.0 ml/min
Temperature: 30°C
Detection: UV280nm

Sample: 1; Doxycycline (10µg)

NACALAI TESQUE, INC

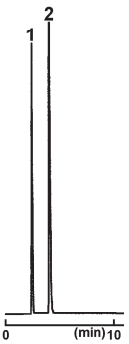
AP-0640

● Doxifluridine

COSMOSIL Application Data

Column: $5C_{18}$-AR-II
Column size: 4.6mmI.D.-150mm
Mobile phase: Methanol/ H_2O = 35/65
Flow rate: 1.0 ml/min
Temperature: 25°C
Detection: UV254nm

Sample: 1; (+)-*5*-Fluoro-*5′*-deoxyuridine [Doxifluridine] (0.5µg)
2; Caffeine (1.0µg)



NACALAI TESQUE, INC

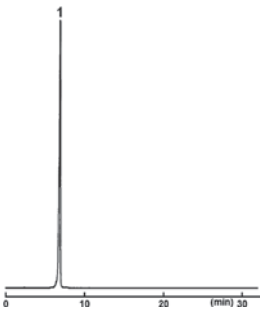
AP-0635

● Doxorubicin Hydrochloride

COSMOSIL Application Data

Column: $5C_{18}$-MS-II
Column size: 4.6mmI.D.-250mm
Mobile phase: Acetonitrile/ 10.4mmol/l Sodium Lauryl Sulfate, 0.14%H_3PO_4=50/50
Flow rate: 1.0 ml/min
Temperature: 25°C
Detection: UV254nm

Sample: 1; Doxorubicin (5.0µg)



NACALAI TESQUE, INC

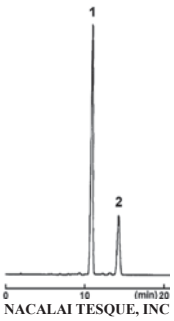
AP-0637

● Tocopherols

COSMOSIL Application Data

Column: $5C_{18}$-MS-II
Column size: 4.6mmI.D.-150mm
Mobile phase: Methanol/ H_2O = 98/2
Flow rate: 1.0 ml/min
Temperature: 30°C
Detection: UV292nm

Sample: 1; α-Tocopherol (20µg)
2; *DL*-α-Tocopherol Acetate (20µg)



NACALAI TESQUE, INC

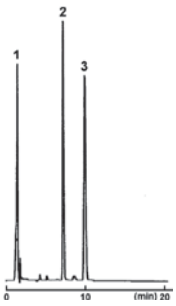
AP-0873

● Tocopherol Nicotinate

COSMOSIL Application Data

Column: $5C_{18}$-MS-II
Column size: 4.6mmI.D.-150mm
Mobile phase: Methanol
Flow rate: 1.0 ml/min
Temperature: 35°C
Detection: UV264nm

Sample: 1; Nicotinic Acid (0.75µg)
2; α-Tocopherol (12.5µg)
3; (±)-α-Tocopherol Nicotinate (2.5µg)



NACALAI TESQUE, INC

AP-0876

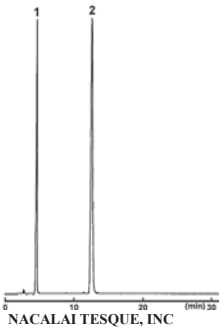
(1) Drugs

● Todralazine Hydrochloride

COSMOSIL Application Data

Column: $5C_{18}$-MS-II
Column size: 4.6mmI.D.-250mm
Mobile phase: Methanol/ 5.4mmol/l Sodium *1*-Heptanesulfonate = 40/60 (pH3.5 with Acetic Acid)
Flow rate: 1.0 ml/min
Temperature: 25°C
Detection: UV240nm

Sample: 1; Phthalic Acid (0.4µg)
2; Todralazine (0.4µg)



NACALAI TESQUE, INC

AP-0879

● Dopamine Hydrochloride

COSMOSIL Application Data

Column: $5C_{18}$-MS-II
Column size: 4.6mmI.D.-250mm
Mobile phase: 200mmol/l Na_2HPO_4 (pH3.0 with Citric Acid)
Flow rate: 1.0 ml/min
Temperature: 25°C
Detection: UV280nm

Sample: 1; Uracil (0.5µg)
2; Dopamine (1.0µg)

NACALAI TESQUE, INC

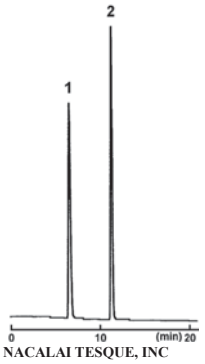
AP-0631

● Dobutamine Hydrochloride

COSMOSIL Application Data

Column: $5C_{18}$-MS-II
Column size: 4.6mmI.D.-250mm
Mobile phase: Methanol/ 20mmol/l Tartrate buffer (pH3.0) = 30/70
Flow rate: 1.0 ml/min
Temperature: 30°C
Detection: UV280nm

Sample: 1; Dobutamine (11µg)
2; Salicylamide (8.5µg)



NACALAI TESQUE, INC

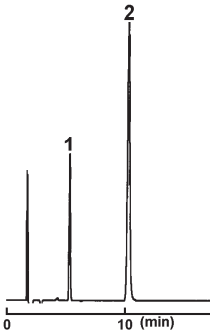
AP-0628

● Tolazamide

COSMOSIL Application Data

Column: 5SL-II
Column size: 4.6mmI.D.-250mm
Mobile phase: Hexane/ Water-saturated Hexane/ Tetrahydrofuran/ Ethanol/ Acetic Acid = 475/ 475/ 20/ 15/ 9
Flow rate: 2.0 ml/min
Temperature: 30°C
Detection: UV254nm

Sample: 1; Tolbutamide (7.5µg)
2; Tolazamide (30µg)



NACALAI TESQUE, INC

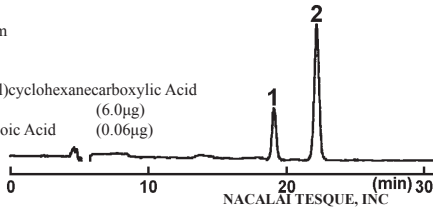
AP-0882

● Tranexamic Acid

COSMOSIL Application Data

Column: $5C_{18}$-PAQ
Column size: 4.6mmI.D.-250mm
Mobile phase: Methanol/ 152mmol/l NaH_2PO_4, 8mmol/l Sodium Lauryl Sulfate, 0.83%Triethylamine(pH2.5 with H_3PO_4) = 40/60
Flow rate: 0.5 ml/min
Temperature: 35°C
Detection: UV220nm

Sample:
1; *trans-4*-(Aminomethyl)cyclohexanecarboxylic Acid [Tranexamic Acid] (6.0µg)
2; *4*-(Aminomethyl)benzoic Acid (0.06µg)



NACALAI TESQUE, INC

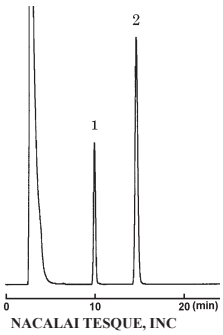
AP-0888

● Triamcinolone

COSMOSIL Application Data

Column: $5C_{18}$-MS-II
Column size: 4.6mmI.D.-150mm
Mobile phase: Acetonitrile/ H_2O = 25/75
Flow rate: 0.5 ml/min
Temperature: 30°C
Detection: UV254nm

Sample: 1; Triamcinolone (1.0µg)
2; Methyl *p*-Hydroxybenzoate (0.4µg)



NACALAI TESQUE, INC

AP-0889

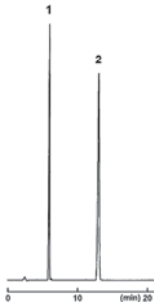
(1) Drugs

● Triamcinolone Acetonide

COSMOSIL Application Data

Column: $5C_{18}$-MS-II
Column size: 4.6mmI.D.-250mm
Mobile phase: Acetonitrile/ H_2O = 35/65
Flow rate: 1.0 ml/min
Temperature: 25°C
Detection: UV240nm

Sample: 1; Prednisolone (0.4µg)
2; Triamcinolone Acetonide (0.8µg)



NACALAI TESQUE, INC

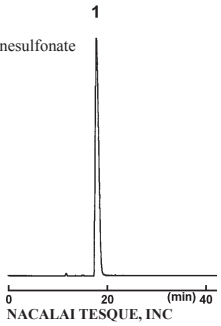
AP-0892

● Trimetazidine Hydrochloride

COSMOSIL Application Data

Column: $5C_{18}$-MS-II
Column size: 4.6mmI.D.-150mm
Mobile phase: A; Methanol/ 14.2mmol/l Sodium *1*-Heptanesulfonate (pH 3.0 with 10% H_3PO_4) = 2/3
B; Methanol
B conc. 5%→25% 50min Linear gradient
Flow rate: 1.0 ml/min
Temperature: 40°C
Detection: UV240nm

Sample: 1; *1*-(*2,3,4*-Trimethoxybenzyl)piperazine [Trimetazidine] (40µg)



NACALAI TESQUE, INC

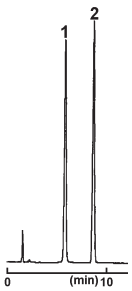
AP-1125

● Trimetazidine Hydrochloride

COSMOSIL Application Data

Column: $5C_{18}$-MS-II
Column size: 4.6mmI.D.-150mm
Mobile phase: Methanol/ 50mmol/l KH_2PO_4 (pH3.0 with H_3PO_4) = 15/85
Flow rate: 1.0 ml/min
Temperature: 40°C
Detection: UV230nm

Sample: 1; *1*-(*2,3,4*-Trimethoxybenzyl)piperazine [Trimetazidine] (0.15µg)
2; *p*-Hydroxybenzoic Acid (0.18µg)



NACALAI TESQUE, INC

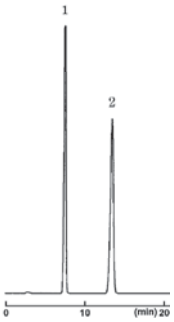
AP-0895

● Tolnaftate

COSMOSIL Application Data

Column: $5C_{18}$-PAQ
Column size: 4.6mmI.D.-150mm
Mobile phase: Methanol/ H_2O = 70/30
Flow rate: 1.0 ml/min
Temperature: 25°C
Detection: UV254nm

Sample: 1; Diphenyl Phthalate (18µg)
2; Tolnaftate (4.0µg)



NACALAI TESQUE, INC

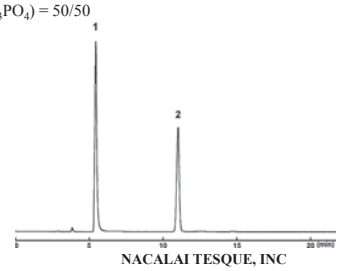
AP-0885

● Domperidone

COSMOSIL Application Data

Column: $5C_8$-MS
Column size: 4.6mmI.D.-250mm
Mobile phase: Methanol/ 15.6mmol/l K_2HPO_4 (pH3.5 with 20mmol/l H_3PO_4) = 50/50
Flow rate: 1.0 ml/min
Temperature: 35°C
Detection: UV287nm

Sample:
1; Domperidone (0.1µg)
2; Ethyl *p*-hydroxybenzoate (0.2µg)



NACALAI TESQUE, INC

AP-1102

● Naphazoline and Chlorpheniramine

COSMOSIL Application Data

Column: $5C_{18}$-AR-II
Column size: 4.6mmI.D.-250mm
Mobile phase: Acetonitrile/ 0.2%Sodium Lauryl Sulfate, 0.1%H_3PO_4 = 50/50
Flow rate: 1.0 ml/min
Temperature: 30°C
Detection: UV254nm

Sample: 1; *o*-Ethoxybenzamide (5.1µg)
2; Naphazoline Nitrate (2.2µg)
3; Chlorpheniramine (4.1µg)

NACALAI TESQUE, INC

AP-0954

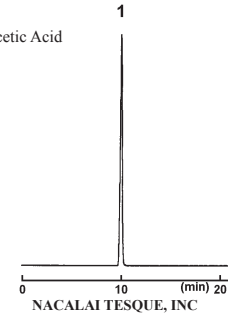
(1) Drugs

● Nabumetone

COSMOSIL Application Data

Column: $5C_{18}$-AR-II
Column size: 4.6mmI.D.-150mm
Mobile phase: Acetonitrile/ Tetrahydrofuran/ 0.1% Acetic Acid = 28/12/60
Flow rate: 1.0 ml/min
Temperature: 40°C
Detection: UV254nm

Sample: 1; Nabumetone (10.0µg)



NACALAI TESQUE, INC

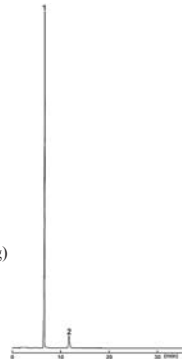
AP-1113

● Nalidixic Acid

COSMOSIL Application Data

Column: $5C_{18}$-AR-II
Column size: 4.6mmI.D.-150mm
Mobile phase: Methanol/ 40mmol/l NaH_2PO_4 (pH2.8 with H_3PO_4) = 40/60
Flow rate: 1.0 ml/min
Temperature: 40°C
Detection: UV260nm

Sample: 1; Methyl *p*-Hydroxybenzoate (0.13 µg)
2; Nalidixic Acid (0.005 µg)



NACALAI TESQUE, INC

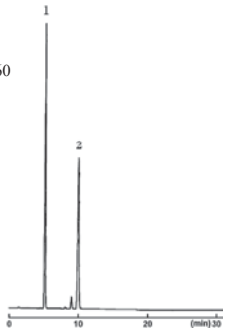
AP-0770

● Nicardipine Hydrochloride

COSMOSIL Application Data

Column: $5C_{18}$-MS-II
Column size: 4.6mmI.D.-150mm
Mobile phase: Acetonitrile/ 0.05%Perchloric Acid = 40/60
Flow rate: 1.0 ml/min
Temperature: 30°C
Detection: UV254nm

Sample: 1; Nicardipine (0.4µg)
2; Nifedipine (0.4µg)



NACALAI TESQUE, INC

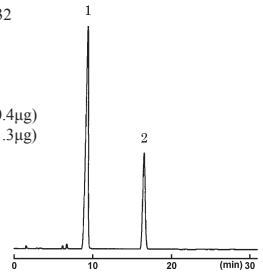
AP-0775

● Nicardipine Hydrochloride

COSMOSIL Application Data

Column: $5C_{18}$-MS-II
Column size: 4.6mmI.D.-150mm
Mobile phase: Methanol/ 10mmol/l KH_2PO_4 = 68/32
Flow rate: 1.0 ml/min
Temperature: 40°C
Detection: UV254nm

Sample: 1; Nicardipine (0.4µg)
2; Di-*n*-butyl Phthalate (1.3µg)



NACALAI TESQUE, INC

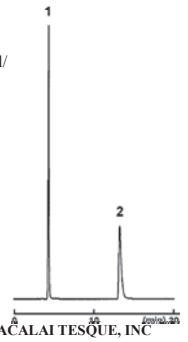
AP-0778

● Nicotinic Acid

COSMOSIL Application Data

Column: $5C_{18}$-AR-II
Column size: 4.6mmI.D.-150mm
Mobile phase: 5mmol/l Sodium *1*-Octanesulfonate-Methanol/ 50mmol/l NaH_2PO_4(pH3.0) = 20/80
Flow rate: 0.5 ml/min
Temperature: 35°C
Detection: UV260nm

Sample: 1; Nicotinic Acid (1.0µg)
2; Caffeine (1.0µg)



NACALAI TESQUE, INC

AP-0788

● Nicotinamide

COSMOSIL Application Data

Column: $5C_{18}$-MS-II
Column size: 4.6mmI.D.-250mm
Mobile phase: Methanol/ 5mmol/l Sodium *1*-Heptanesulfonate = 30/70
Flow rate: 0.5 ml/min
Temperature: 25°C
Detection: UV254nm

Sample: 1; Nicotinic Acid (4.0µg)
2; Nicotinamide (0.8µg)

NACALAI TESQUE, INC

AP-0784

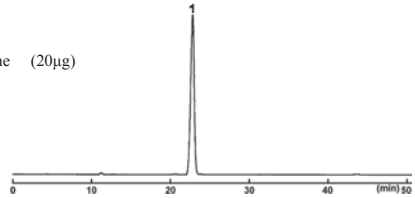
(1) Drugs

● Nicergolin

COSMOSIL Application Data

Column: $5C_{18}$-MS-II
Column size: 4.6mmI.D.-250mm
Mobile phase: Methanol/Acetonitrile/ 50mmol/l KH_2PO_4 (pH7.0 with Triethylamine) = 35/30/35
Flow rate: 1.0 ml/min
Temperature: 40°C
Detection: UV288nm

Sample: 1; Nicergoline (20µg)



NACALAI TESQUE, INC

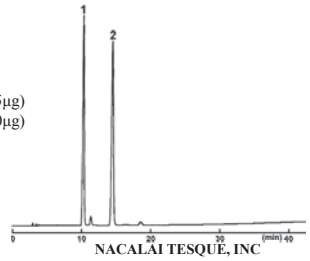
AP-0781

● Nitrendipine

COSMOSIL Application Data

Column: $5C_{18}$-MS-II
Column size: 4.6mmI.D.-150mm
Mobile phase: Acetonitrile/Tetrahydrofuran/H_2O = 20/24/56
Flow rate: 0.5 ml/min
Temperature: 30°C
Detection: UV254nm

Sample:
1; *n*-Propyl *p*-Hydroxybenzoate (0.15µg)
2; Nitrendipine (0.50µg)



NACALAI TESQUE, INC

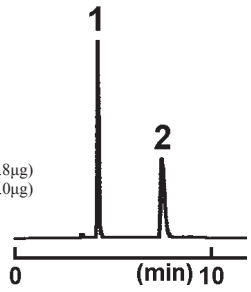
AP-0790

● Neostigmine Methylsulfate

COSMOSIL Application Data

Column: $5C_{18}$-MS-II
Column size: 4.6mmI.D.-150mm
Mobile phase: Acetonitrile/ 20mmol/l NaH_2PO_4 (pH3.0 with H_3PO_4), 5mmol/l Sodium *1*-Pentanesulfonate = 11/89
Flow rate: 1.0 ml/min
Temperature: 25°C
Detection: UV259nm

Sample: 1; Dimethylaminophenol (0.8µg)
2; Neostigmine Methyl Sulfate (5.0µg)



NACALAI TESQUE, INC

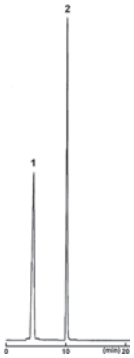
AP-0772

● Baclofen

COSMOSIL Application Data

Column: $5C_{18}$-AR-II
Column size: 4.6mmI.D.-250mm
Mobile phase: Methanol/ 0.11%Acetic acid = 60/40
Flow rate: 0.5 ml/min
Temperature: 25°C
Detection: UV268nm

Sample: 1; Baclofen (5.0µg)
2; Methyl *p*-Hydroxybenzoate (0.063µg)



NACALAI TESQUE, INC

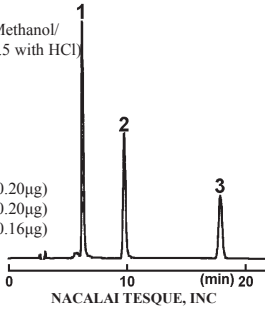
AP-0477

● Haloperidol

COSMOSIL Application Data

Column: $5C_{18}$-AR-II
Column size: 4.6mmI.D.-150mm
Mobile phase: 3.5mmol/l Sodium Lauryl Sulfate-Methanol/ 10mmol/l *tri* - Sodium Citrate(pH3.5 with HCl) = 75/25
Flow rate: 0.5 ml/min
Temperature: 40°C
Detection: UV220nm

Sample:
1; *4*-(*4*-Chlorophenyl)-*4*-Hydroxypiperidine (0.20µg)
2; Haloperidol (0.20µg)
3; Biphenyl (0.16µg)



NACALAI TESQUE, INC

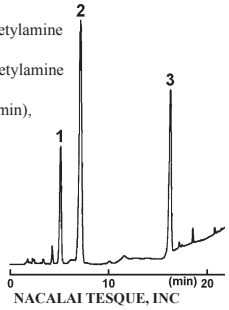
AP-0680

● Vancomycin Hydrochloride

COSMOSIL Application Data

Column: $5C_{18}$-MS-II
Column size: 4.6mmI.D.-250mm
Mobile phase: A; Acetonitrile/Tetrahydrofuran/ 0.2%Trietylamine (pH3.2 with H_3PO_4) = 7/1/92
B; Acetonitrile/Tetrahydrofuran/ 0.2%Trietylamine (pH3.2 with H_3PO_4) = 29/1/70
B conc. 0%(0-12min), 0→100%(12→20min), 100%(20-22min)
Flow rate: 1.5 ml/min
Temperature: 25°C
Detection: UV280nm

Sample: 1; Similar compound 1
2; Vancomycin (5.0µg)
3; Similar compound 2



NACALAI TESQUE, INC

AP-0901

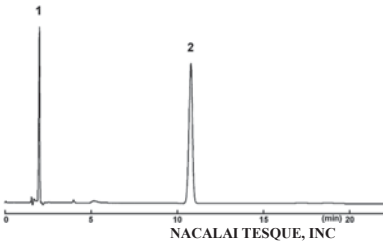
(1) Drugs

● Bisoprolol Fumarate

COSMOSIL Application Data

Column: $5C_8$-MS
Column size: 4.6mmI.D.-150mm
Mobile phase: Acetonitrile/ 30mmol/l KH_2PO_4 (pH2.5 with H_3PO_4) = 20/80
Flow rate: 1.0 ml/min
Temperature: 40°C
Detection: UV225nm

Sample: 1; Fumaric Acid
2; Bisoprolol



NACALAI TESQUE, INC

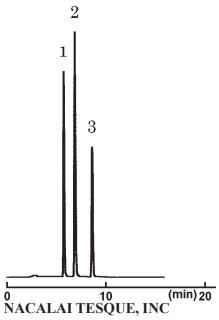
AP-1090

● Hydrochlorothiazide

COSMOSIL Application Data

Column: $5C_{18}$-AR-II
Column size: 4.6mmI.D.-250mm
Mobile phase: Acetonitrile/ 100mmol/l NaH_2PO_4 (pH3.0 with H_3PO_4) =20/80
Flow rate: 1.0 ml/min
Temperature: 25°C
Detection: UV254nm

Sample:
1; *4*-Amino-*6*-chloro-*1*,*3*-benzenedisulfonamide (1.0µg)
2; Hydrochlorothiazide (3.0µg)
3; *p*-Aminoacetophenone (3.6µg)



NACALAI TESQUE, INC

AP-0683

● Hydrocortisone

COSMOSIL Application Data

Column: 5SL-II
Column size: 4.6mmI.D.-250mm
Mobile phase: Chloroform/Methanol/Acetic Acid = 1000/20/1
Flow rate: 2.0 ml/min
Temperature: 30°C
Detection: UV254nm

Sample: 1; Prednisone (0.9µg)
2; Hydrocortisone (2.0µg)

NACALAI TESQUE, INC

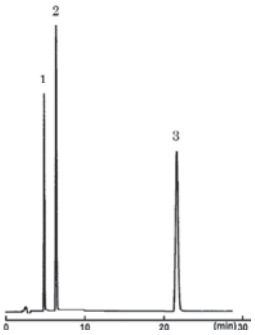
AP-0685

● Hydrocortisone Succinate

COSMOSIL Application Data

Column: $5C_{18}$-MS-II
Column size: 4.6mmI.D.-250mm
Mobile phase: Acetonitrile/ 40mmol/l CH_3COONa (pH4.0 with Acetic Acid) = 40/60
Flow rate: 1.0 ml/min
Temperature: 25°C
Detection: UV254nm

Sample:
1; Hydrocortisone (0.4µg)
2; Hydrocortisone *21*-Hemisuccinate (1.0µg)
3; Butyl *p*-Hydroxybenzoate (0.3µg)



NACALAI TESQUE, INC

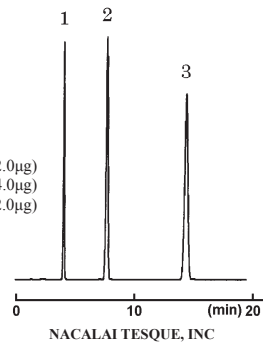
AP-0689

● Hydrocortisone Acetate

COSMOSIL Application Data

Column: $5C_{18}$-MS-II
Column size: 4.6mmI.D.-250mm
Mobile phase: Acetonitrile/ H_2O = 45/55
Flow rate: 1.0 ml/min
Temperature: 30°C
Detection: UV254nm

Sample: 1; Hydrocortisone (2.0µg)
2; Hydrocortisone Acetate (4.0µg)
3; Benzyl *p*-Hydroxybenzoate (2.0µg)



NACALAI TESQUE, INC

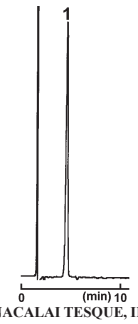
AP-0686

● Hypromellose Phthalate (impurity)

COSMOSIL Application Data

Column: $5C_{18}$-AR-II
Column size: 4.6mmI.D.-250mm
Mobile phase: Acetonitrile/ 100mmol/l Cyanoacetic Acid = 15/85
Flow rate: 2.0 ml/min
Temperature: 30°C
Detection: UV235nm

Sample: 1; Phthalic Acid (0.5µg)



NACALAI TESQUE, INC

AP-0933

(1) Drugs

● Piperacillin Sodium

COSMOSIL Application Data

Column: $5C_{18}$-MS-II
Column size: 4.6mmI.D.-150mm
Mobile phase: Acetonitrile/ 50mmol/l Acetic Acid, 25mmol/l Triethylamine = 21/79
Flow rate: 1.0 ml/min
Temperature: 30°C
Detection: UV254nm

Sample: 1; Ampicillin (10µg)
2; Acetanilide (0.75µg)
3; Piperacillin (2.5µg)

NACALAI TESQUE, INC

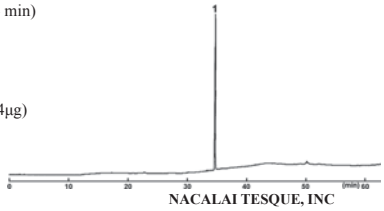
AP-0799

● Piperacillin Sodium

COSMOSIL Application Data

Column: $5C_{18}$-MS-II
Column size: 4.6mmI.D.-150mm
Mobile phase: A; Acetonitrile/H_2O/ 200mmol/l KH_2PO_4 = 4/45/1
B; Acetonitrile/H_2O/ 200mmol/l KH_2PO_4 = 25/24/1
B conc. 0%(0-7 min), 0→17%(7→13 min), 17%(13-25 min), 17→80%(25→40min), 80%(40-44 min)
Flow rate: 1.0 ml/min
Temperature: 30°C
Detection: UV220nm

Sample: 1; Piperacillin (0.4µg)



NACALAI TESQUE, INC

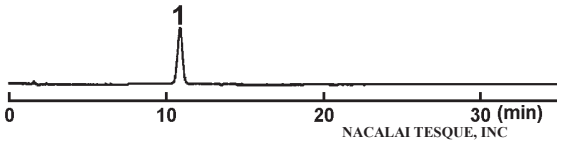
AP-0802

● Pimaricin

COSMOSIL Application Data

Column: $5C_{18}$-MS-II
Column size: 4.6mmI.D.-150mm
Mobile phase: 0.1%Ammonium Acetate-Methanol/Tetrahydrofuran/H_2O = 44/2/47
Flow rate: 1.0 ml/min
Temperature: 40°C
Detection: UV303nm

Sample: 1; Pimaricin (0.02µg)



NACALAI TESQUE, INC

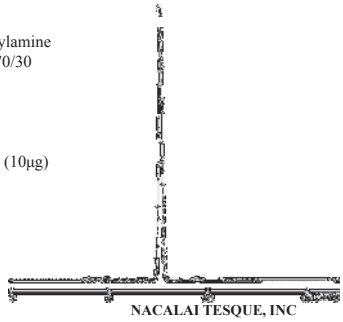
AP-0796

● Vincristine Sulfate

COSMOSIL Application Data

Column: $5C_8$-MS
Column size: 4.6mmI.D.-250mm
Mobile phase: Methanol/ 1.67% Diethylamine ($pH7.5$ with H_3PO_4) = 70/30
Flow rate: 1.0 ml/min
Temperature: 30°C
Detection: UV297nm

Sample: 1; Vincristine Sulfate (10µg)



NACALAI TESQUE, INC

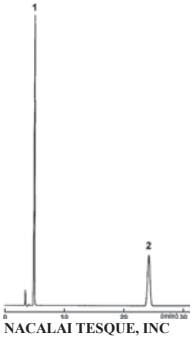
AP-1128

● Famotidine

COSMOSIL Application Data

Column: $5C_{18}$-AR-II
Column size: 4.6mmI.D.-150mm
Mobile phase: Acetonitrile/Methanol/ 10mmol/l Sodium *1*-Heptanesulfonate ($pH3.0$ with Acetic Acid) = 19/3/78
Flow rate: 0.5 ml/min
Temperature: 25°C
Detection: UV254nm

Sample: 1; Famotidine (0.5µg)
2; Methyl *p*-Hydroxybenzoate (0.8µg)



NACALAI TESQUE, INC

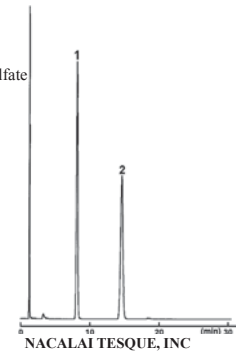
AP-0651

● Scopolamine Butyl Bromide

COSMOSIL Application Data

Column: $5C_8$-MS
Column size: 4.6mmI.D.-150mm
Mobile phase: Methanol/ 19mmol/l Sodium Lauryl Sulfate = 68/37($pH3.6$ with H_3PO_4)
Flow rate: 1.0 ml/min
Temperature: 30°C
Detection: UV210nm

Sample:
1; Scopolamine (2.0µg)
2; (-)-Scopolamine *n*-Butyl Bromide (2.0µg)



NACALAI TESQUE, INC

AP-0852

(1) Drugs

● Bufexamac

COSMOSIL Application Data

Column: $5C_{18}$-AR-II
Column size: 4.6mmI.D.-150mm
Mobile phase: Methanol/Acetonitrile/
13.6mmol/l Sodium *1*-Octane Sulfonate
0.94%Acetic Acid, 1.9mmol/l EDTA
= 24/24/52
Flow rate: 1.0 ml/min
Temperature: 30°C
Detection: UV275nm

Sample:
1; Bufexamac (2 μg)
2; *4,5*-Diphenylimidazole (0.16 μg)

NACALAI TESQUE, INC

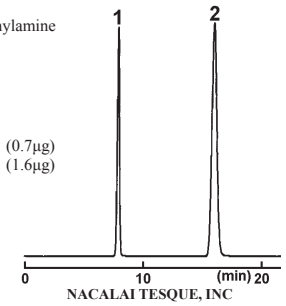
AP-0522

● Pravastatin

COSMOSIL Application Data

Column: $5C_{18}$-MS-II
Column size: 4.6mmI.D.-150mm
Mobile phase: Methanol/H_2O/Acetic Acid/Triethylamine
= 500/500/1/1
Flow rate: 1.0 ml/min
Temperature: 25°C
Detection: UV238nm

Sample:
1; Ethyl *p*-Hydroxybenzoate (0.7μg)
2; Pravastatin (1.6μg)



NACALAI TESQUE, INC

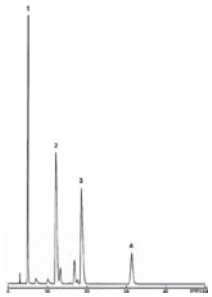
AP-0811

● Flavin Adenine Dinucleotide Sodium

COSMOSIL Application Data

Column: $5C_{18}$-MS-II
Column size: 4.6mmI.D.-250mm
Mobile phase: Methanol/ 0.2%KH_2PO_4 = 20/80
Flow rate: 1.0 ml/min
Temperature: 35°C
Detection: UV260nm

Sample:
1; Adenosine (1.0μg)
2; Flavin Adenine Dinucleotide [FAD] (4.0μg)
3; Flavin Mononucleotide [FMN] (4.0μg)
4; Vitamin B_2 [Riboflavin] (1.0μg)



NACALAI TESQUE, INC

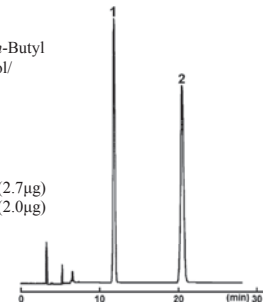
AP-0653

● Fluoxymesterone

COSMOSIL Application Data

Column: 5SL-II
Column size: 4.6mmI.D.-250mm
Mobile phase: *n*-Butyl Chloride/Water-saturated *n*-Butyl
Chloride/Tetrahydrofuran/Methanol/
Acetic Acid = 95/95/14/7/6
Flow rate: 1.0 ml/min
Temperature: 30°C
Detection: UV254nm

Sample:
1; Fluoxymesterone (2.7μg)
2; *6α*-Methylprednisolone (2.0μg)



NACALAI TESQUE, INC

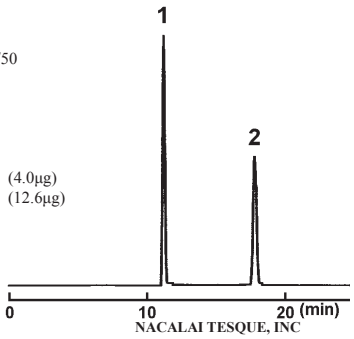
AP-0669

● Fluocinonide

COSMOSIL Application Data

Column: $5C_{18}$-MS-II
Column size: 4.6mmI.D.-250mm
Mobile phase: Acetonitrile/ H_2O = 50/50
Flow rate: 1.0 ml/min
Temperature: 40°C
Detection: UV254nm

Sample:
1; Fluocinolone Acetonide *21*-Acetate (4.0μg)
2; Propyl Benzoate (12.6μg)



NACALAI TESQUE, INC

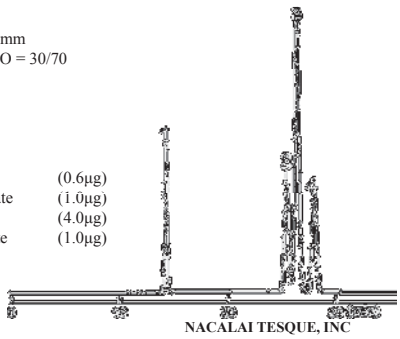
AP-0663

● Fluocinolone Acetonide

COSMOSIL Application Data

Column: $5C_{18}$-AR-II
Column size: 4.6mmI.D.-250mm
Mobile phase: Acetonitrile/ H_2O = 30/70
Flow rate: 1.0 ml/min
Temperature: 40°C
Detection: UV254nm

Sample:
1; Ethyl *p*-Hydroxybenzoate (0.6μg)
2; Isopropyl *p*-Hydroxybenzoate (1.0μg)
3; Fluocinolone Acetonide (4.0μg)
4; *n*-Propyl *p*-Hydroxybenzoate (1.0μg)



NACALAI TESQUE, INC

AP-0660

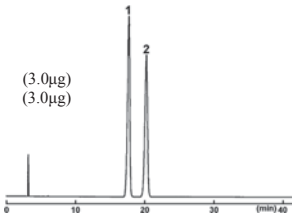
(1) Drugs

● Fluocinolone Acetonide

COSMOSIL Application Data

Column: 5SL-II
Column size: 4.6mmI.D.-250mm
Mobile phase: Water-saturated Chloroform/Methanol/ Acetic Acid = 200/3/2
Flow rate: 1.0 ml/min
Temperature: 30°C
Detection: UV254nm

Sample: 1; Triamcinolone Acetonide (3.0µg)
2; Fluocinolone Acetonide (3.0µg)



NACALAI TESQUE, INC

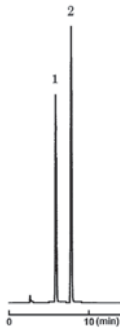
AP-0662

● Fluorometholone

COSMOSIL Application Data

Column: $5C_{18}$-MS-II
Column size: 4.6mmI.D.-250mm
Mobile phase: Methanol/ H_2O = 70/30
Flow rate: 1.0 ml/min
Temperature: 35°C
Detection: UV254nm

Sample: 1; Fluorometholone (0.20µg)
2; Butyl *p*-Hydroxybenzoate (0.16µg)



NACALAI TESQUE, INC

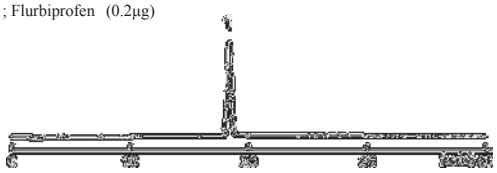
AP-0666

● Flurbiprofen

COSMOSIL Application Data

Column: $5C_{18}$-AR-II
Column size: 4.6mmI.D.-150mm
Mobile phase: Acetonitrile/ 7.7%H_3PO_4 = 35/65
Flow rate: 1.0 ml/min
Temperature: 30°C
Detection: UV254nm

Sample: 1; Flurbiprofen (0.2µg)



NACALAI TESQUE, INC

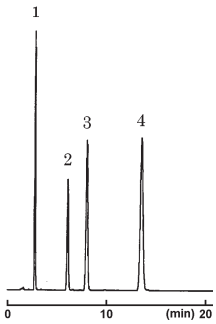
AP-0671

● Prednisolone Acetate

COSMOSIL Application Data

Column: $5C_{18}$-MS-II
Column size: 4.6mmI.D.-150mm
Mobile phase: Acetonitrile/ H_2O = 40/60
Flow rate: 1.0 ml/min
Temperature: 25°C
Detection: UV254nm

Sample: 1; Prednisolone (1.5µg)
2; Prednisolone *21*-Acetate (1.0µg)
3; Cortisone-*21*-Acetate (1.5µg)
4; Butyl *p*-Hydroxybenzoate (0.5µg)



NACALAI TESQUE, INC

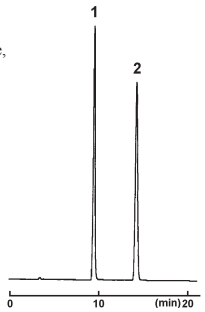
AP-0814

● Procaine Hydrochloride

COSMOSIL Application Data

Column: $5C_{18}$-MS-II
Column size: 4.6mmI.D.-150mm
Mobile phase: Methanol/ 0.1%Sodium *1*-Pentanesulfonate, 50mmol/l KH_2PO_4(pH3.0 with H_3PO_4) = 20/80
Flow rate: 0.5 ml/min
Temperature: 40°C
Detection: UV254nm

Sample: 1; Procaine (1.25µg)
2; Caffeine (1.25µg)



NACALAI TESQUE, INC

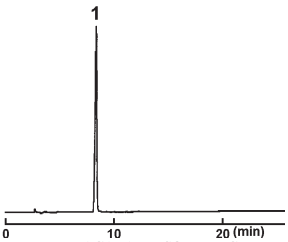
AP-0817

● Procaterol Hydrochloride

COSMOSIL Application Data

Column: $5C_{18}$-AR-II
Column size: 4.6mmI.D.-250mm
Mobile phase: Methanol/ 5mmol/l Sodium *1*-Pentanesulfonate/ Acetic Acid = 23/76/1
Flow rate: 1.0 ml/min
Temperature: 40°C
Detection: UV254nm

Sample: 1; Procaterol (0.06µg)



NACALAI TESQUE, INC

AP-0821

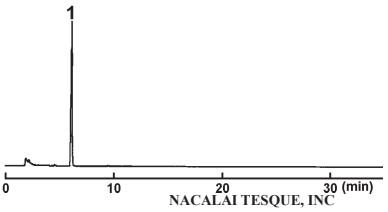
(1) Drugs

● Propranolol Hydrochloride

COSMOSIL Application Data

Column: $5C_{18}$-MS-II
Column size: 4.6mmI.D.-250mm
Mobile phase: Acetonitrile/ 12mmol/l Sodium Lauryl Sulfate, 0.7mmol/l Tetra-*n*-butylammonium Phosphate/ Sulfuric Acid = 550/450/1(pH3.3 with NaOH)
Flow rate: 1.0 ml/min
Temperature: 25°C
Detection: UV292nm

Sample:
1; Propranolol (0.08µg)



NACALAI TESQUE, INC

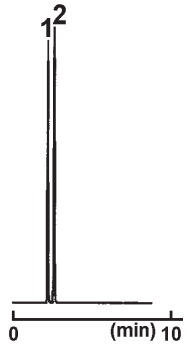
AP-0823

● Flopropione

COSMOSIL Application Data

Column: $5C_{18}$-AR-II
Column size: 4.6mmI.D.-150mm
Mobile phase: Acetonitrile/ 1.2%H_3PO_4 = 57/43
Flow rate: 1.0 ml/min
Temperature: 35°C
Detection: UV267nm

Sample: 1; *2',4',6'*-Trihydroxypropiophenone [Flopropione] (0.40µg)
2; Ethyl *4*-Hydroxybenzoate (0.26µg)



NACALAI TESQUE, INC

AP-0657

● Bromhexine Hydrochloride

COSMOSIL Application Data

Column: $5C_{18}$-MS-II
Column size: 4.6mmI.D.-150mm
Mobile phase: Acetonitrile/ 7.3mmol/l KH_2PO_4 (pH7.0 with NaOH) = 80/20
Flow rate: 2.0 ml/min
Temperature: 40°C
Detection: UV245nm

Sample: 1; Bamethane (25µg)
2; Bromhexine (1.25µg)

NACALAI TESQUE, INC

AP-0518

● Beclometasone Dipropionate

COSMOSIL Application Data

Column: $5C_{18}$-MS-II
Column size: 4.6mmI.D.-150mm
Mobile phase: Acetonitrile/ H_2O = 60/40
Flow rate: 1.0 ml/min
Temperature: 25°C
Detection: UV254nm

Sample: 1; Beclomethasone Dipropionate (1.6µg)
2; Testosterone Propionate (0.8µg)

NACALAI TESQUE, INC

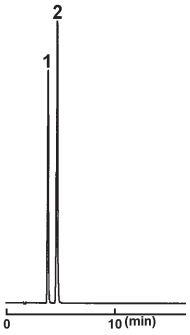
AP-0479

● Bezafibrate

COSMOSIL Application Data

Column: $5C_{18}$-AR-II
Column size: 4.6mmI.D.-150mm
Mobile phase: Methanol/ 0.1%Acetic Acid = 9/4
Flow rate: 1.0 ml/min
Temperature: 30°C
Detection: UV230nm

Sample: 1; *4*-Chlorobenzoic Acid (0.55µg)
2; Bezafibrate (1.05µg)



NACALAI TESQUE, INC

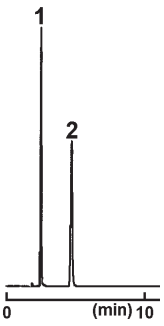
AP-0510

● Bezafibrate

COSMOSIL Application Data

Column: $5C_{18}$-AR-II
Column size: 4.6mmI.D.-150mm
Mobile phase: Methanol/ 0.1%Acetic Acid = 9/4
Flow rate: 1.0 ml/min
Temperature: 30°C
Detection: UV230nm

Sample: 1; *p*-Nitrophenol (0.32µg)
2; Bezafibrate (0.44µg)



NACALAI TESQUE, INC

AP-0513

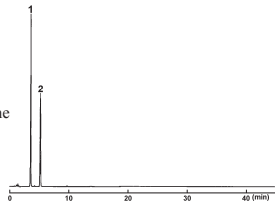
(1) Drugs

● Betahistine Mesilate

COSMOSIL Application Data

Column: $5C_{18}$-MS-II
Column size: 4.6mmI.D.-150mm
Mobile phase: 8mmol/l Sodium Lauryl Sulfate-Acetonitrile/ 2%Acetic Acid, 0.5%Diethylamine = 37/63
Flow rate: 1.0 ml/min
Temperature: 35°C
Detection: UV261nm

Sample: 1; *2*-Vinylpyridine (0.16µg)
2; *2*-(*2*-Methylaminoethyl)pyridine [Betahistine] (0.16µg)



NACALAI TESQUE, INC

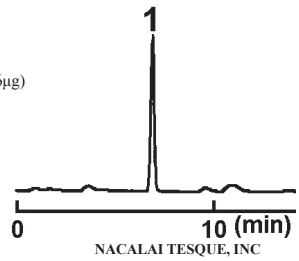
AP-0488

● Betamethasone

COSMOSIL Application Data

Column: $5C_{18}$-MS-II
Column size: 4.6mmI.D.-150mm
Mobile phase: Methanol/ H_2O = 60/40
Flow rate: 1.0 ml/min
Temperature: 25°C
Detection: UV241nm

Sample: 1; Betamethasone (0.06µg)



NACALAI TESQUE, INC

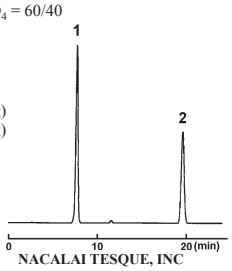
AP-0497

● Betamethasone Sodium Phosphate

COSMOSIL Application Data

Column: $5C_{18}$-MS-II
Column size: 4.6mmI.D.-250mm
Mobile phase: Methanol / 5mmol/l Tetra-*n*-butylammonium Bromide 8.9mmol/l Na_2HPO_4,50.7mmol/l KH_2PO_4 = 60/40
Flow rate: 1.0 ml/min
Temperature: 25°C
Detection: UV254nm

Sample: 1; Betamethasone *21*-Phosphate (1.0µg)
2; Butyl *p*-Hydroxybenzoate (0.2µg)



NACALAI TESQUE, INC

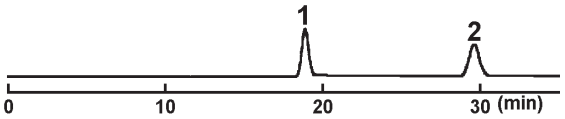
AP-0494

● Betamethasone Valerate

COSMOSIL Application Data

Column: $5C_{18}$-MS-II
Column size: 4.6mmI.D.-150mm
Mobile phase: Methanol/ H_2O = 65/35
Flow rate: 1.0 ml/min
Temperature: 30°C
Detection: UV254nm

Sample: 1; Betamethasone *17*-Valerate (1.5µg)
2; Beclomethasone Dipropionate (1.5µg)



NACALAI TESQUE, INC

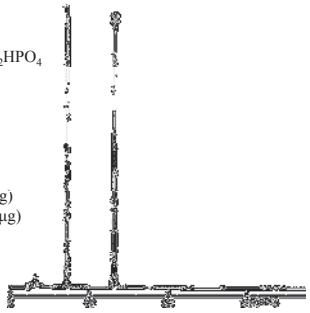
AP-0503

● Benzylpenicillin Potassium

COSMOSIL Application Data

Column: $5C_{18}$-MS-II
Column size: 4.6mmI.D.-250mm
Mobile phase: Acetonitrile/ 50mmol/l $(NH_4)_2HPO_4$ = 24/76 (pH8 with H_3PO_4)
Flow rate: 1.0 ml/min
Temperature: 25°C
Detection: UV254nm

Sample:
1; Penicillin G (4.0µg)
2; Methyl *p*-Hydroxybenzoate (0.06µg)



NACALAI TESQUE, INC

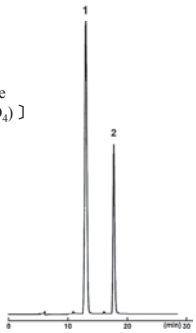
AP-0482

● Calcium Folinate

COSMOSIL Application Data

Column: $5C_{18}$-MS-II
Column size: 4.6mmI.D.-250mm
Mobile phase: [H_2O/Acetonitrile/Methanol/ 40%Tetra-*n*-butylammonium Hydroxide = 760/200/8.6/9.4 (pH7.5 with NaH_2PO_4)] →1000(with H_2O)
Flow rate: 0.5 ml/min
Temperature: 30°C
Detection: UV254nm

Sample: 1; Calcium Folinate (2.8µg)
2; Folic Acid (0.8µg)



NACALAI TESQUE, INC

AP-0524

(1) Drugs

● Calcium Folinate

COSMOSIL Application Data

Column: $5C_{18}$-MS-II
Column size: 4.6mmI.D.-150mm
Mobile phase: Methanol/ 8mmol/l Na_2HPO_4/ Tetrabutylammonium Hydroxide = 110/385/4 (pH 7.5 with H_3PO_4)
Flow rate: 1.0 ml/min
Temperature: 45°C
Detection: UV254nm

Sample:
1; Calcium Folinate (2.0µg)
2; Folic Acid (2.0µg)

NACALAI TESQUE, INC

AP-1093

● Mitomycin C

COSMOSIL Application Data

Column: 5PE-MS
Column size: 4.6mmI.D.-250mm
Mobile phase: Methanol/ 20mmol/l Ammonium Acetate, 0.025%Acetic Acid = 40/60
Flow rate: 1.0 ml/min
Temperature: 25°C
Detection: UV365nm

Sample:
1; Mitomycin C (5.0µg)
2; Ethyl Vanillin
[*3*-Ethoxy-*4*-Hydroxybenzaldehyde] (75µg)

NACALAI TESQUE, INC

AP-0768

● Mizoribine

COSMOSIL Application Data

Column: $5C_{18}$-PAQ
Column size: 4.6mmI.D.-250mm
Mobile phase: 0.067% H_3PO_4 aq.
Flow rate: 1.0 ml/min
Temperature: 30°C
Detection: UV279nm

Sample:
1; Mizoribine (1.0µg)

NACALAI TESQUE, INC

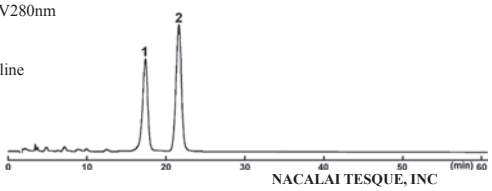
AP-1108

● Minocycline Hydrochloride

COSMOSIL Application Data

Column: $5C_8$-MS
Column size: 4.6mmI.D.-150mm
Mobile phase: 2.8%Ammonium Oxalate/*N,N*-dimethylformamide/ 100mmol/l EDTA = 11/5/4 (pH6.2 with Tetrabutylammonium Hydroxide)
Flow rate: 1.0 ml/min
Temperature: 30°C
Detection: UV280nm

Sample:
1; *4-epi*-Minocycline
2; Minocycline



NACALAI TESQUE, INC

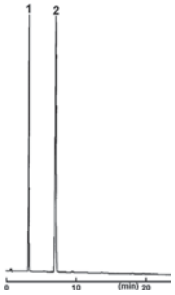
AP-0767

● Mexiletine Hydrochloride

COSMOSIL Application Data

Column: $5C_8$-MS
Column size: 4.6mmI.D.-150mm
Mobile phase: Acetonitrile/ 14.4mmol/l Sodium Lauryl Sulfate, 25mmol/l NaH_2PO_4 = 21/30
Flow rate: 2.0 ml/min
Temperature: 30°C
Detection: UV210nm

Sample:
1; β-Phenylethylamine (0.6µg)
2; *1*-(2,*6*-Dimethylphenoxy)-*2*-Propanamine [Mexiletine] (1.0µg)



NACALAI TESQUE, INC

AP-0766

● Mecobalamin

COSMOSIL Application Data

Column: $5C_{18}$-PAQ
Column size: 4.6mmI.D.-250mm
Mobile phase: 100mmol/l Sodium *1*-Hexanesulfonate, Acetonitrile/ 20mmol/l Phosphate buffer(pH3.5) = 20/80
Flow rate: 0.5 ml/min
Temperature: 40°C
Detection: UV266nm

Sample:
1; Vitamin B_{12} [Cyanocobalamin] (0.5µg)
2; Hydroxocobalamin Acetate (0.5µg)
3; Mecobalamin (1.0µg)

NACALAI TESQUE, INC

AP-0738

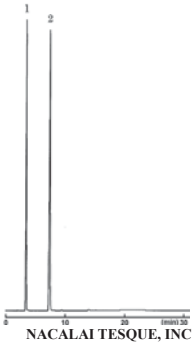
(1) Drugs

● Meticrane

COSMOSIL Application Data

Column: $5C_{18}$-MS-II
Column size: 4.6mmI.D.-150mm
Mobile phase: Acetonitrile/ H_2O = 15/85
Flow rate: 1.0 ml/min
Temperature: 40°C
Detection: UV230nm

Sample: 1; Caffeine (0.2µg)
2; Meticrane (0.2µg)



NACALAI TESQUE, INC

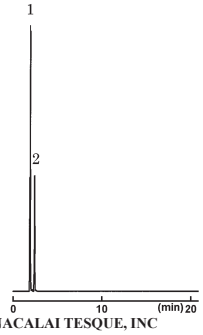
AP-0754

● Meticrane

COSMOSIL Application Data

Column: $5C_{18}$-MS-II
Column size: 4.6mmI.D.-150mm
Mobile phase: Acetonitrile/ H_2O = 50/50
Flow rate: 1.0 ml/min
Temperature: 40°C
Detection: UV230nm

Sample: 1; Meticrane (0.4µg)
2; Methyl *p*-Hydroxybenzoate (0.4µg)



NACALAI TESQUE, INC

AP-0757

● Methotrexate

COSMOSIL Application Data

Column: $5C_{18}$-MS-II
Column size: 4.6mmI.D.-250mm
Mobile phase: Acetonitrile/ 200mmol/l Na_2HPO_4
(pH6.0 with 100mmol/l Citric Acid) = 10/90
Flow rate: 1.0 ml/min
Temperature: 30°C
Detection: UV302nm

Sample: 1; Folic Acid (1.0µg)
2; Methotrexate (1.0µg)

NACALAI TESQUE, INC

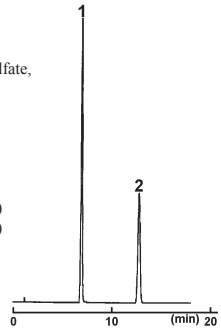
AP-0751

● Metformin Hydrochloride

COSMOSIL Application Data

Column: $5C_{18}$-AR-II
Column size: 4.6mmI.D.-150mm
Mobile phase: Acetonitrile/ 4.5mmol/l Sodium Lauryl Sulfate,
0.04%H_3PO_4 = 38/62
Flow rate: 1.0 ml/min
Temperature: 40°C
Detection: UV235nm

Sample: 1; *1,1*-Dimethylbiguanide
[Metformin] (0.50µg)
2; Isobutyl *p*-Hydroxybenzoate (0.95µg)



NACALAI TESQUE, INC

AP-0761

● Metronidazole

COSMOSIL Application Data

Column: $5C_{18}$-MS-II
Column size: 4.6mmI.D.-150mm
Mobile phase: Methanol/ H_2O = 20/80
Flow rate: 1.0 ml/min
Temperature: 30°C
Detection: UV320nm

Sample: 1; Metronidazole (0.24µg)

NACALAI TESQUE, INC

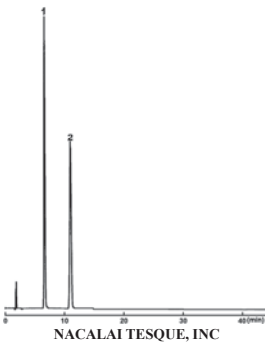
AP-0763

● Menatetrenone

COSMOSIL Application Data

Column: $5C_{18}$-MS-II
Column size: 4.6mmI.D.-150mm
Mobile phase: Methanol
Flow rate: 1.0 ml/min
Temperature: 40°C
Detection: UV270nm

Sample: 1; Menatetrenone (0.6µg)
2; Vitamin K_1 (0.6µg)



NACALAI TESQUE, INC

AP-0742

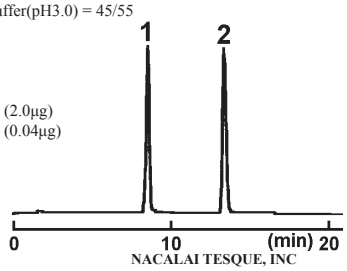
(1) Drugs

● Mepivacaine Hydrochloride

COSMOSIL Application Data

Column: $5C_{18}$-AR-II
Column size: 4.6mmI.D.-150mm
Mobile phase: 10mmol/l Sodium Lauryl Sulfate-Acetonitrile/ 20mmol/l Phosphate Buffer(pH3.0) = 45/55
Flow rate: 1.0 ml/min
Temperature: 30°C
Detection: UV254nm

Sample:
1; Mepivacaine (2.0µg)
2; Benzophenone (0.04µg)



NACALAI TESQUE, INC

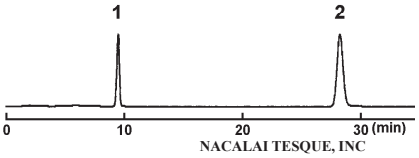
AP-0746

● Meropenem

COSMOSIL Application Data

Column: $5C_{18}$-AR-II
Column size: 4.6mmI.D.-150mm
Mobile phase: Methanol/ 0.1%Triethylamine (pH5.0 with H_3PO_4) = 10/90
Flow rate: 1.0 ml/min
Temperature: 25°C
Detection: UV220nm

Sample:
1; Meropenem (2.5µg)
2; Benzyl Alcohol (1.5µg)



NACALAI TESQUE, INC

AP-0749

● Ubidecarenone

COSMOSIL Application Data

Column: $5C_{18}$-PAQ
Column size: 4.6mmI.D.-150mm
Mobile phase: Methanol/ Ethanol = 50/50
Flow rate: 0.5 ml/min
Temperature: 35°C
Detection: UV275nm

Sample:
1; Coenzyme Q10 [Ubidecarenone] (2.5µg)

NACALAI TESQUE, INC

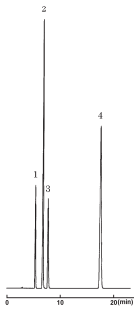
AP-0900

● Iodine, Salicylic Acid and Phenol

COSMOSIL Application Data

Column: $5C_{18}$-MS-II
Column size: 4.6mmI.D.-250mm
Mobile phase: Methanol/ 100mmol/l Phosphate Buffer(pH7.0) = 25/75
Flow rate: 1.0 ml/min
Temperature: 30°C
Detection: UV270nm

Sample:
1; Benzoic Acid (2.0µg)
2; Theophylline (0.5µg)
3; Salicylic Acid (2.0µg)
4; Phenol (2.0µg)



NACALAI TESQUE, INC

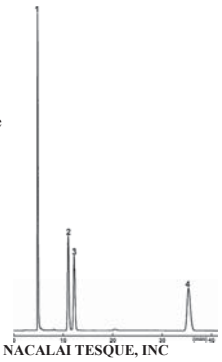
AP-0935

● Latamoxef Sodiums

COSMOSIL Application Data

Column: $5C_{18}$-MS-II
Column size: 4.6mmI.D.-150mm
Mobile phase: Methanol/ 51mmol/l KH_2PO_4, 9mmol/l Na_2HPO_4, 5mmol/l Tetra-*n*-Butylammonium Bromide = 25/75
Flow rate: 1.0 ml/min
Temperature: 25°C
Detection: UV254nm

Sample:
1; *5*-Mercapto-*1*-Methyltetrazole (1.65µg)
2; Latamoxef (isomer 1)
3; Latamoxef (isomer 2)
4; *m*-Cresol (7.5µg)



NACALAI TESQUE, INC

AP-0711

● Latamoxef Sodiums

COSMOSIL Application Data

Column: $5C_{18}$-PAQ
Column size: 4.6mmI.D.-150mm
Mobile phase: Methanol/ 100mmol/l Ammonium Acetate = 5/95
Flow rate: 1.0 ml/min
Temperature: 25°C
Detection: UV254nm

Sample:
1; Latamoxef (isomer 1)
2; Latamoxef (isomer 2)

NACALAI TESQUE, INC

AP-0716

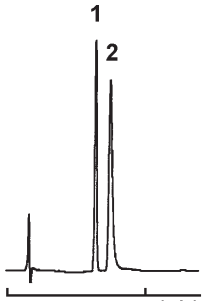
(1) Drugs

● Liothyronine Sodium

COSMOSIL Application Data

Column: $5C_{18}$-PAQ
Column size: 4.6mmI.D.-250mm
Mobile phase: Methanol/ H_2O = 60/40
Flow rate: 1.0 ml/min
Temperature: 25°C
Detection: UV220nm

Sample: 1; *n*-Propyl *p*-Hydroxybenzoate (0.14µg)
2; *3,3',5*-Triiodo-*L*-thyronine (0.08µg)



NACALAI TESQUE, INC

AP-0728

● Lisinopril

COSMOSIL Application Data

Column: $5C_{18}$-PAQ
Column size: 4.6mmI.D.-150mm
Mobile phase: A; 25mmol/l NaH_2PO_4
B; Acetonitrile/ 25mmol/l NaH_2PO_4 = 40/60
B conc. 10→50%(0→10min), 50%(10-25min)
Flow rate: 1.5 ml/min
Temperature: 60°C
Detection: UV215nm

Sample: 1; Lisinopril (0.45µg)
2; Caffeine (0.015µg)

NACALAI TESQUE, INC

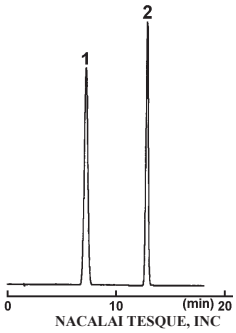
AP-0732

● Lisinopril

COSMOSIL Application Data

Column: $5C_{18}$-MS-II
Column size: 4.6mmI.D.-150mm
Mobile phase: Acetonitrile/ 25mmol/l NaH_2PO_4 = 5/95
Flow rate: 1.0 ml/min
Temperature: 60°C
Detection: UV215nm

Sample: 1; Lisinopril (2.0µg)
2; Caffeine (0.5µg)



NACALAI TESQUE, INC

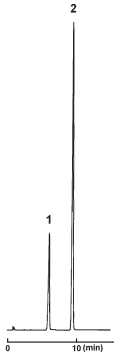
AP-0733

● Lidocaine

COSMOSIL Application Data

Column: $5C_{18}$-AR-II
Column size: 4.6mmI.D.-150mm
Mobile phase: 10mmol/l Sodium Lauryl Sulfate-Acetonitrile/ 20mmol/l Phosphate Buffer(pH3.0) =45/55
Flow rate: 1.5 ml/min
Temperature: 25°C
Detection: UV254nm

Sample: 1; Lidocaine (8.5µg)
2; Benzophenone (0.25µg)



NACALAI TESQUE, INC

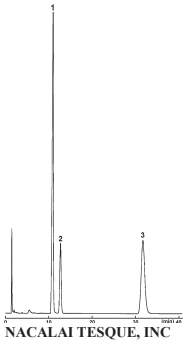
AP-0724

● Ritodrine Hydrochloride

COSMOSIL Application Data

Column: $5C_{18}$-AR-II
Column size: 4.6mmI.D.-150mm
Mobile phase: Methanol/ 71mmol/l $(NH_4)_2HPO_4$, 7.8mmol/l Sodium *1*-Heptanesulfonate = 30/70(pH3.0 with H_3PO_4)
Flow rate: 1.0 ml/min
Temperature: 25°C
Detection: UV220nm

Sample: 1; Ritodrine
2; *threo*-Ritodrine
3; by-product



NACALAI TESQUE, INC

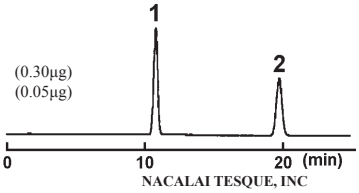
AP-0835

● Ritodrine Hydrochloride

COSMOSIL Application Data

Column: $5C_{18}$-AR-II
Column size: 4.6mmI.D.-150mm
Mobile phase: Methanol/ 71mmol/l $(NH_4)_2HPO_4$, 7.8mmol/l Sodium *1*-Heptanesulfonate = 30/70(pH3.0 with H_3PO_4)
Flow rate: 1.0 ml/min
Temperature: 25°C
Detection: UV274nm

Sample:
1; Ritodrine (0.30µg)
2; Methyl *p*-Hydroxybenzoate (0.05µg)



NACALAI TESQUE, INC

AP-0838

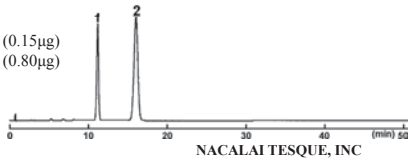
(1) Drugs

● Rifampicin

COSMOSIL Application Data

Column: $5C_8$-MS
Column size: 4.6mmI.D.-150mm
Mobile phase: 20mmol/l Citric Acid, 11mmol/l Sodium Perchlorate-Acetonitrile/H_2O/ 1mol/l KH_2PO_4, 55mmol/l H_3PO_4(pH3.1) = 7/11/2
Flow rate: 2.0 ml/min
Temperature: 25°C
Detection: UV254nm

Sample:
1; Butyl *p*-Hydroxybenzoate (0.15µg)
2; Rifampicin (0.80µg)



NACALAI TESQUE, INC

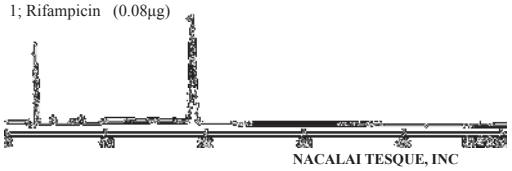
AP-0832

● Rifampicin

COSMOSIL Application Data

Column: $5C_8$-MS
Column size: 4.6mmI.D.-250mm
Mobile phase: Acetonitrile/ 15mmol/l Sodium Perchlorate, 28mmol/l Citric Acid, 17mmol/l NaH_2PO_4 = 45/55
Flow rate: 1.0 ml/min
Temperature: 25°C
Detection: UV254nm

Sample: 1; Rifampicin (0.08µg)



NACALAI TESQUE, INC

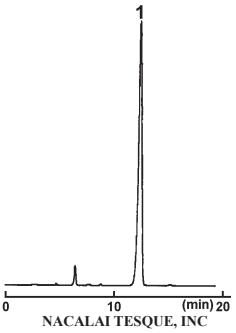
AP-0833

● Lincomycin Hydrochloride

COSMOSIL Application Data

Column: $5C_8$-MS
Column size: 4.6mmI.D.-250mm
Mobile phase: Acetonitrile/Methanol/ 1.35%H_3PO_4 (pH6.0 with Ammonia) = 15/15/78
Flow rate: 1.0 ml/min
Temperature: 46°C
Detection: UV210nm

Sample: 1; Lincomycin (20µg)



NACALAI TESQUE, INC

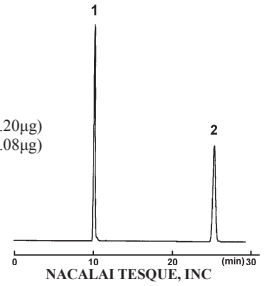
AP-0729

● Reserpine

COSMOSIL Application Data

Column: $5C_{18}$-AR-II
Column size: 4.6mmI.D.-250mm
Mobile phase: Acetonitrile/ 50mmol/l KH_2PO_4 (pH3.0 with H_3PO_4) = 45/55
Flow rate: 0.5 ml/min
Temperature: 40°C
Detection: UV268nm

Sample: 1; Reserpine (0.20µg)
2; Butyl *p*-Hydroxybenzoate (0.08µg)



NACALAI TESQUE, INC

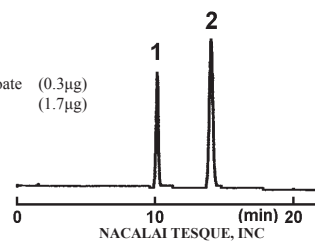
AP-0830

● Levallorphan Tartrate

COSMOSIL Application Data

Column: $5C_{18}$-AR-II
Column size: 4.6mmI.D.-150mm
Mobile phase: Acetonitrile/ 7mmol/l Sodium Lauryl Sulfate, 0.1%H_3PO_4(pH3.0 with NaOH) = 40/60
Flow rate: 1.0 ml/min
Temperature: 40°C
Detection: UV280nm

Sample: 1; Isobutyl *4*-Hydroxybenzoate (0.3µg)
2; Levallorphan (1.7µg)



NACALAI TESQUE, INC

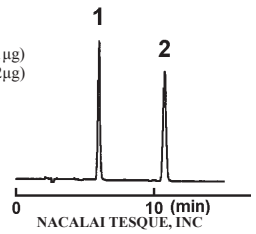
AP-0718

● Levothyroxine

COSMOSIL Application Data

Column: $5C_{18}$-AR-II
Column size: 4.6mmI.D.-250mm
Mobile phase: Methanol/ 0.15%H_3PO_4 =67/33
Flow rate: 1.0 ml/min
Temperature: 25°C
Detection: UV230nm

Sample: 1; Levothyroxine [*L*-Thyroxine] (0.1µg)
2; Ethinylestradiol (0.2µg)



NACALAI TESQUE, INC

AP-0721

(1) Drugs

● Roxatidine Acetate Hydrochloride

COSMOSIL Application Data

Column: $5C_{18}$-MS-II
Column size: 4.6mmI.D.-150mm
Mobile phase: Acetonitrile/ H_2O/ Triethylamine/ Acetic Acid = 60/340/2/1
Flow rate: 1.0 ml/min
Temperature: 40°C
Detection: UV274nm

Sample: 1; Roxatidine Acetate (4.2µg)

NACALAI TESQUE, INC

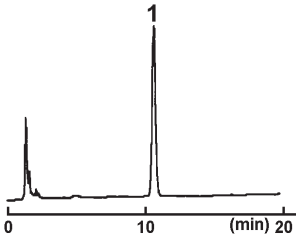
AP-0840

● Roxithromycin

COSMOSIL Application Data

Column: $5C_{18}$-PAQ
Column size: 4.6mmI.D.-150mm
Mobile phase: Acetonitrile/ 0.2mmol/l $(NH_4)H_2PO_4$ (pH5.3 with NaOH) =30/70
Flow rate: 1.0 ml/min
Temperature: 30°C
Detection: UV205nm

Sample: 1; Roxithromycin (1.0µg)



NACALAI TESQUE, INC

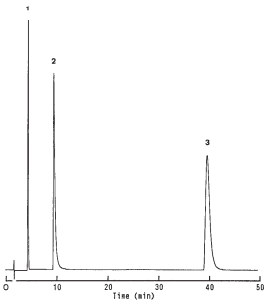
AP-0845

● Tricyclic Drugs

COSMOSIL Application Data

Column: $5C_{18}$-MS-II
Column size: 4.6mmI.D.-150mm
Mobile phase: Methanol/ 20mmol/l Phosphate Buffer(pH7) = 60/40
Flow rate: 1.0 ml/min
Temperature: 30°C
Detection: UV254nm, 0.2AUFS

Sample: 1; Carbamazepine
2; Desipramine
3; Imipramine



NACALAI TESQUE, INC

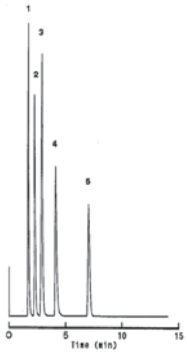
AP-0066

● Bronchodilators

COSMOSIL Application Data

Column: $5C_{18}$-AR-II
Column size: 4.6mmI.D.-150mm
Mobile phase: Methanol/ H_2O = 30/70
Flow rate: 1.0 ml/min
Temperature: 30°C
Detection: UV254nm, 1.0AUFS

Sample: 1; Uracil (0.5µg)
2; Theobromine (1.5µg)
3; Theophylline (2.0µg)
4; Caffeine (2.0µg)
5; Phenol (0.8µg)



NACALAI TESQUE, INC

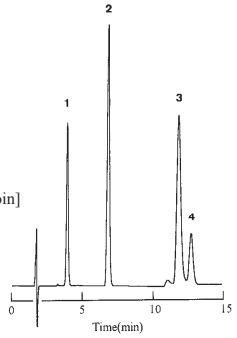
AP-0076

● Antiarrhythmic Drugs

COSMOSIL Application Data

Column: $5C_{18}$-MS-II
Column size: 4.6mmI.D.-150mm
Mobile phase: Methanol/ 20mmol/l Phosphate Buffer(pH7) = 60/40
Flow rate: 1.0 ml/min
Temperature: 30°C
Detection: UV254nm, 0.05AUFS

Sample: 1; Phenytoin [*5,5*-Diphenylhydantoin]
2; Ketamine
3; Quinidine
4; Lidocaine



NACALAI TESQUE, INC

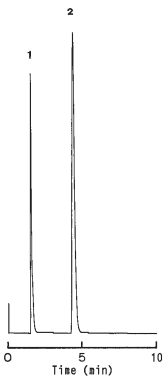
AP-0067

● Antiarrhythmic Drugs

COSMOSIL Application Data

Column: $5C_{18}$-AR-II
Column size: 4.6mmI.D.-150mm
Mobile phase: Methanol/ 20mmol/l Phosphate Buffer(pH2) = 10/90
Flow rate: 1.0 ml/min
Temperature: 30°C
Detection: UV254nm, 0.2AUFS

Sample: 1; Procainamide (0.5µg)
2; *N*-Acetylprocainamide (0.5µg)



NACALAI TESQUE, INC

AP-0069

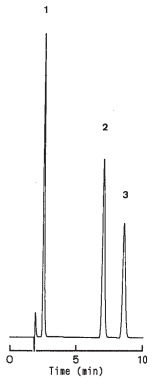
(1) Drugs

● Antiepileptics

COSMOSIL Application Data

Column: $5C_{18}$-MS-II
Column size: 4.6mmI.D.-150mm
Mobile phase: Methanol/ 20mmol/l Phosphate Buffer(pH7) = 50/50
Flow rate: 1.0 ml/min
Temperature: 30°C
Detection: UV254nm, 0.5AUFS

Sample:
1; Barbital (7.59μg)
2; Phenytoin [*5,5*-Diphenylhydantoin] (10.38μg)
3; Carbamazepine (1.02μg)



NACALAI TESQUE, INC

AP-0077

● Analgesics

COSMOSIL Application Data

Column: $5C_{18}$-AR-II
Column size: 4.6mmI.D.-150mm
Mobile phase: Methanol / 20mmol/l Phosphoric Acid = 40/60
Flow rate: 1.0 ml/min
Temperature: 30°C
Detection: UV254nm, 0.5AUFS

Sample:
1; *4*-Aminoantipyrine (0.74μg)
2; *p*-Acetamidophenol (2.97μg)
3; Antipyrine (0.85μg)
4; Acetylsalicylic Acid〔Aspyrine〕
5; Phenacetin (0.54μg)
6; Salicylic acid

NACALAI TESQUE, INC

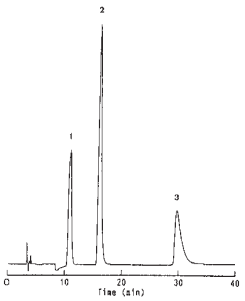
AP-0073

● Analgesics

COSMOSIL Application Data

Column: 5SL-II
Column size: 4.6mmI.D.-250mm
Mobile phase: Ethyl Acetate/Hexane =1/1
Flow rate: 1.0 ml/min
Temperature: 30°C
Detection: UV254nm, 0.2AUFS

Sample:
1; Acetanilide (1.0μg)
2; Phenacetin (1.0μg)
3; *p*-Acetamidophenol (1.0μg)



NACALAI TESQUE, INC

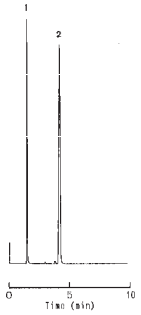
AP-0074

● Histamine H1–Receptor Blockers

COSMOSIL Application Data

Column: $5C_{18}$-MS-II
Column size: 4.6mmI.D.-150mm
Mobile phase: Acetonitrile/ 10mmol/l SDS, 0.1% Phosphoric Acid = 60/40
Flow rate: 1.0 ml/min
Temperature: 30°C
Detection: UV254nm

Sample:
1; Uracil
2; Diphenhydramine



NACALAI TESQUE, INC

AP-0078

● Histamine H2–Receptor Blockers

COSMOSIL Application Data

Column: $5C_{18}$-MS-II
Column size: 4.6mmI.D.-150mm
Mobile phase: Methanol/ 20mmol/l Phosphate buffer(pH7) = 30/70
Flow rate: 1.0 ml/min
Temperature: 30°C
Detection: UV254nm, 0.2AUFS

Sample:
1; Famotidine (1.3μg)
2; Cimetidine (33.5μg)
3; Ranitidine (1.6μg)

NACALAI TESQUE, INC

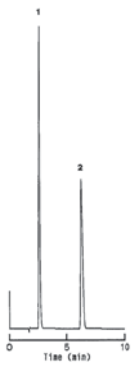
AP-0079

● Antihyperlipidemic Drugs

COSMOSIL Application Data

Column: $5C_{18}$-MS-II
Column size: 4.6mmI.D.-150mm
Mobile phase: Methanol / 20mmol/l Phosphoric Acid = 10/90
Flow rate: 1.0 ml/min
Temperature: 30°C
Detection: UV254nm, 0.5AUFS

Sample:
1; Hydralazine (1.0μg)
2; Todralazine (1.0μg)



NACALAI TESQUE, INC

AP-0080

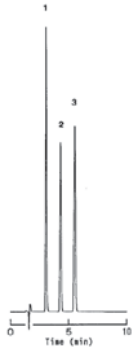
(1) Drugs

● Profens

COSMOSIL Application Data

Column: $5C_{18}$-MS-II
Column size: 4.6mmI.D.-150mm
Mobile phase: Acetonitrile/ 20mmol/l Acetic Acid = 60/40
Flow rate: 1.0 ml/min
Temperature: 30°C
Detection: UV220nm, 1.0AUFS

Sample: 1; Ketoprofen (1.60µg)
2; Ibuprofen (1.69µg)
3; Flurbiprofen (1.57µg)



NACALAI TESQUE, INC

AP-0081

● Cardiac Glycosides

COSMOSIL Application Data

Column: $5C_{18}$-AR-II
Column size: 4.6mmI.D.-150mm
Mobile phase: Methanol/ H_2O = 70/30
Flow rate: 1.0 ml/min
Temperature: 30°C
Detection: UV230nm, 0.32AUFS

Sample: 1; Digitoxigenin (2.5µg)
2; Digitoxin (5.0µg)

NACALAI TESQUE, INC

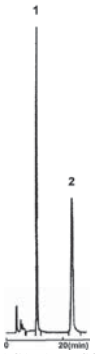
AP-0083

● Anticancer Drugs

COSMOSIL Application Data

Column: Sugar-D
Column size: 4.6mmI.D.-150mm
Mobile phase: Acetonitrile/H_2O = 80/20
Flow rate: 0.3 ml/min
Temperature: Room temperature
Detection: UV226nm

Sample: 1; *cis*-Platin (CDDP) (1.46µg)
2; Guanosine (0.50µg)



NACALAI TESQUE, INC

Data courtesy of Dr.K.Kofuji,Hokuriku University

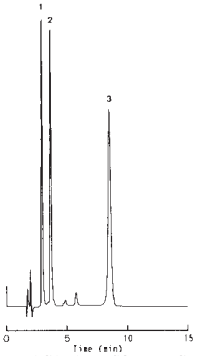
AP-0380

● Tetracyclines Antibiotics

COSMOSIL Application Data

Column: $5C_{18}$-MS-II
Column size: 4.6mmI.D.-150mm
Mobile phase: Methanol/ 20mmol/l Phosphate buffer(pH3) = 20/80
Flow rate: 1.0 ml/min
Temperature: 20°C
Detection: UV254nm

Sample: 1; Oxytetracycline
2; Tetracycline
3; Chlortetracycline



NACALAI TESQUE, INC

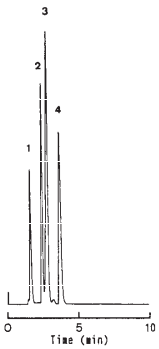
AP-0085

● Penicillin Antibiotics

COSMOSIL Application Data

Column: $5C_{18}$-MS-II
Column size: 4.6mmI.D.-150mm
Mobile phase: Methanol/ 20mmol/l Phosphate Buffer(pH7) = 50/50
Flow rate: 1.0 ml/min
Temperature: 30°C
Detection: UV235nm, 0.2AUFS

Sample: 1; Carbenicillin (1.5µg)
2; Ampicillin (3.0µg)
3; Methicillin (1.5µg)
4; Penicillin G (3.0µg)



NACALAI TESQUE, INC

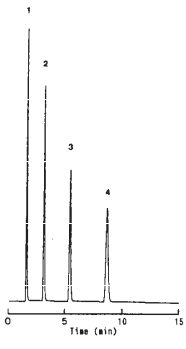
AP-0086

● Quinolone Antimicrobials

COSMOSIL Application Data

Column: $5C_{18}$-MS-II
Column size: 4.6mmI.D.-150mm
Mobile phase: Methanol/ 20mmol/l Phosphate Buffer(pH3) = 55/45
Flow rate: 1.0 ml/min
Temperature: 30°C
Detection: UV270nm, 0.16AUFS

Sample: 1; Ofloxacin (0.39µg)
2; Oxolinic Acid (0.08µg)
3; Flumequine (1.08µg)
4; Piromidic Acid (0.08µg)



NACALAI TESQUE, INC

AP-0087

(1) Drugs

● Nitrofuran Antimicrobials

COSMOSIL Application Data

Column: $5C_{18}$-AR-II
Column size: 4.6mmI.D.-150mm
Mobile phase: Methanol / 20mmol/l Phosphoric Acid = 20/80
Flow rate: 1.0 ml/min
Temperature: 30°C
Detection: UV260nm, 0.5AUFS

Sample: 1; Nitrofurantoin (1.0µg)
2; Nitrofurazone (1.0µg)

NACALAI TESQUE, INC

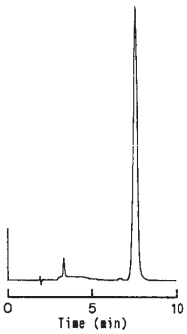
AP-0089

● Streptomycin Sulfate

COSMOSIL Application Data

Column: $5C_{18}$-MS-II
Column size: 4.6mmI.D.-150mm
Mobile phase: Acetonitrile/ 5mmol/l Sodium *1*-Hexanesulfonate, 20mmol/l KH_2PO_4 = 10/90
Flow rate: 1.0 ml/min
Temperature: 30°C
Detection: UV205nm, 0.2AUFS

Sample: Streptomycin Sulfate (5.0µg)



NACALAI TESQUE, INC

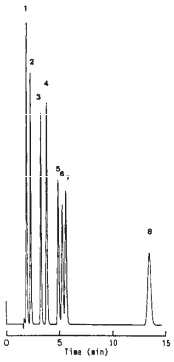
AP-0091

● Sulfa Drugs

COSMOSIL Application Data

Column: $5C_{18}$-MS-II
Column size: 4.6mmI.D.-150mm
Mobile phase: Methanol / 20mmol/l Phosphoric Acid = 30/70
Flow rate: 1.0 ml/min
Temperature: 30°C
Detection: UV230nm, 0.5AUFS

Sample: 1; Sulfaisomidin (0.24µg)
2; Sulfathiazole (0.24µg)
3; Sulfamethazine (0.24µg)
4; Sulfamethoxypyridazine (0.24µg)
5; Sulfamethoxazole (0.24µg)
6; Sulfachloropyridazine (0.24µg)
7; Sulfamonomethoxine (0.24µg)
8; Sulfadimethoxine (0.24µg)



NACALAI TESQUE, INC

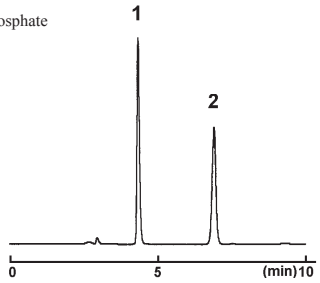
AP-0090

● Allantoin

COSMOSIL Application Data

Column: HILIC
Column size: 4.6mmI.D.-250mm
Mobile phase: Acetonitrile/ 10mmol/l Phosphate Buffer(pH7.0) = 70/30
Flow rate: 1.0 ml/min
Temperature: 30°C
Detection: UV210nm

Sample: 1; Allantoin
2; Allantoic Acid



NACALAI TESQUE, INC

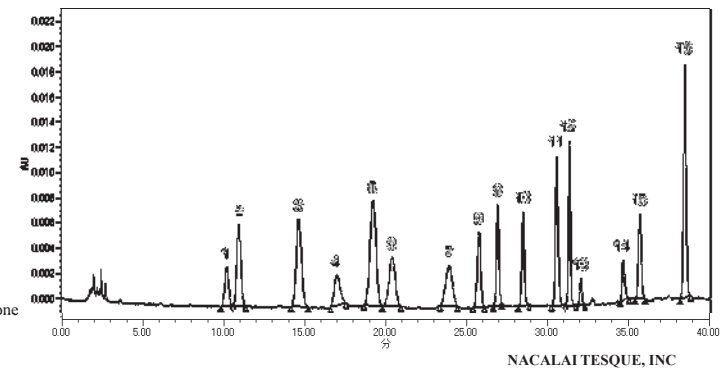
AP-1060

● Tonic Drug

COSMOSIL Application Data

Column: 5PBB-R
Column size: 4.6mmI.D.-150mm
Mobile phase: A: Acetonitrile/ 50mmol/l NaH_2PO_4 = 20/80(pH2.5 with H_3PO_4)
B: Acetonitrile/ 50mmol/l NaH_2PO_4 = 80/20(pH2.5 with H_3PO_4)
B conc. 15→20% (0-20min), 20→100% (20-35min), 100%(35-40min), 15%(40-55min)
Flow rate: 1.0 ml/min
Temperature: 40°C
Detection: UV290nm
Sample:

1; Vardenafil
2; Hydroxyhongdenafil
3; Hongdenafil
4; Thioquinapiperifil
5; Hydroxyhomosildenafil
6; Sildenafil
7; Homosildenafil
8; Aildenafil
9; Udenafil
10; Aminotadalafil
11; Acetil Acid
12; Tadalafil
13; Xanthoanthrafil
14; Thiodenafil
15; Thioaildenafil
16; Imidazosagatriadinone



NACALAI TESQUE, INC

Data courtesy of a customer

AP-1192

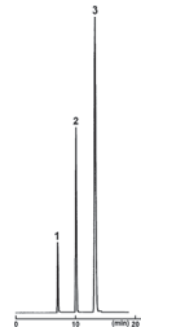
(2) Crude Drugs

● Bearberry Leaf

COSMOSIL Application Data

Column: $5C_{18}$-PAQ
Column size: 4.6mmI.D.-250mm
Mobile phase: Methanol/ 1mmol/l HCl = 5/95
Flow rate: 1.0 ml/min
Temperature: 30°C
Detection: UV280nm

Sample: 1; Arbutin (5.0µg)
2; Hydroquinone (5.0µg)
3; Gallic Acid (5.0µg)



NACALAI TESQUE, INC

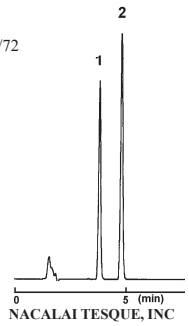
AP-0906

● Scutellaria Root

COSMOSIL Application Data

Column: $5C_{18}$-AR-II
Column size: 4.6mmI.D.-150mm
Mobile phase: Acetonitrile/ 0.68% Phosphoric Acid = 28/72
Flow rate: 1.0 ml/min
Temperature: 50°C
Detection: UV277nm

Sample: 1; Baicalin (0.1µg)
2; Methyl *p*-Hydroxybenzoate (0.2µg)



NACALAI TESQUE, INC

AP-1134

● Phellodendron Bark

COSMOSIL Application Data

Column: $5C_{18}$-PAQ
Column size: 4.6mmI.D.-250mm
Mobile phase: Acetonitrile/ 5.9mmol/l Sodium Lauryl Sulfate, 25mmol/l KH_2PO_4 = 50/50
Flow rate: 1.0 ml/min
Temperature: 40°C
Detection: UV345nm

Sample: 1; Palmatine (1.0µg)
2; Berberine (1.0µg)

NACALAI TESQUE, INC

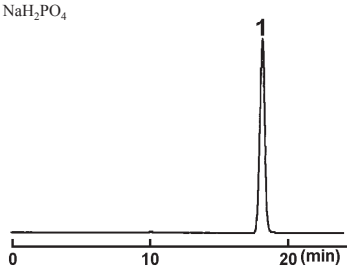
AP-0964

● Pueraria Root

COSMOSIL Application Data

Column: $5C_{18}$-MS-II
Column size: 4.6mmI.D.-150mm
Mobile phase: Acetonitrile/ 50mmol/l NaH_2PO_4 = 10/90
Flow rate: 0.5 ml/min
Temperature: 40°C
Detection: UV250nm

Sample: 1; Puerarin (1.3µg)



NACALAI TESQUE, INC

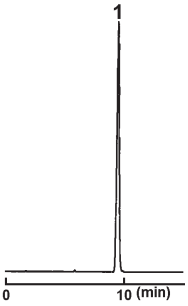
AP-0969

● Kamishoyosan Extract

COSMOSIL Application Data

Column: $5C_{18}$-AR-II
Column size: 4.6mmI.D.-150mm
Mobile phase: Acetonitrile/ 0.11%H_3PO_4 = 10/90
Flow rate: 1.0 ml/min
Temperature: 40°C
Detection: UV240nm

Sample: 1; Geniposide (1.2µg)



NACALAI TESQUE, INC

AP-0942

● Glycyrrhiza

COSMOSIL Application Data

Column: $5C_{18}$-AR-II
Column size: 4.6mmI.D.-250mm
Mobile phase: Acetonitrile/ 2%H_3PO_4 = 40/60
Flow rate: 1.0 ml/min
Temperature: 30°C
Detection: UV254nm

Sample: 1; Glycyrrhizic Acid〔Glycyrrhizin〕 (5.0µg)
2; *n*-Propyl *p*-Hydroxybenzoate (1.0µg)

NACALAI TESQUE, INC

AP-0927

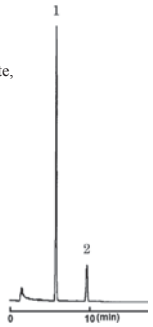
(2) Crude Drugs

● Dried Yeast

COSMOSIL Application Data

Column: $5C_{18}$-PAQ
Column size: 4.6mmI.D.-150mm
Mobile phase: Acetonitrile/ 9.2mmol/l Sodium *1*-Octanesulfonate, 20mmol/l KH_2PO_4(pH3.5 with H_3PO_4) = 20/80
Flow rate: 1.0 ml/min
Temperature: 40°C
Detection: UV254nm

Sample: 1; Vitamin B_1 [Thiamine] (0.10µg)
2; Phenacetin (0.06µg)



NACALAI TESQUE, INC

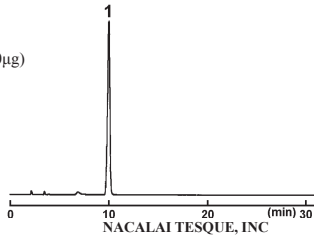
AP-0919

● Keishibukuryougan Extract

COSMOSIL Application Data

Column: $5C_{18}$-MS-II
Column size: 4.6mmI.D.-150mm
Mobile phase: Methanol/ 50mmol/l NaH_2PO_4 = 1/5
Flow rate: 0.8 ml/min
Temperature: 45°C
Detection: UV210nm

Sample: 1; *D*-(-)-Amygdalin (2.0µg)



NACALAI TESQUE, INC

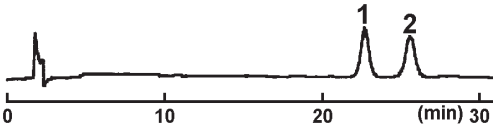
AP-0944

● Red Ginseng and Ginseng

COSMOSIL Application Data

Column: $5C_{18}$-PAQ
Column size: 4.6mmI.D.-150mm
Mobile phase: Acetonitrile/ H_2O = 20/80
Flow rate: 1.0 ml/min
Temperature: 30°C
Detection: UV203nm

Sample: 1; Ginsenoside Rg1 (2.5µg)
2; Ginsenoside Re (2.5µg)



NACALAI TESQUE, INC

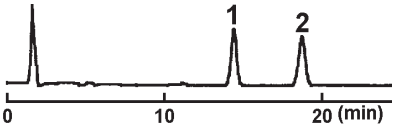
AP-0922

● Red Ginseng and Ginseng

COSMOSIL Application Data

Column: $5C_{18}$-MS-II
Column size: 4.6mmI.D.-150mm
Mobile phase: Acetonitrile/ H_2O = 30/70
Flow rate: 1.0 ml/min
Temperature: 40°C
Detection: UV203nm

Sample: 1; Ginsenoside Rb1 (2.5µg)
2; Ginsenoside Rc (2.5µg)



NACALAI TESQUE, INC

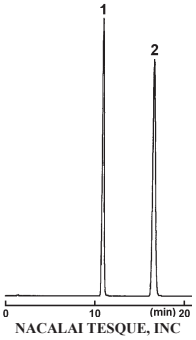
AP-0923

● Magnolia Bark

COSMOSIL Application Data

Column: $5C_{18}$-AR-II
Column size: 4.6mmI.D.-150mm
Mobile phase: Acetonitrile/ 2%Acetic Acid = 50/50
Flow rate: 1.0 ml/min
Temperature: 30°C
Detection: UV289nm

Sample: 1; Honokiol (1.0µg)
2; Magnolol (1.0µg)



NACALAI TESQUE, INC

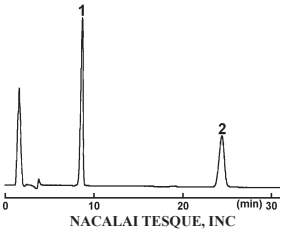
AP-0948

● Bupleurum Root

COSMOSIL Application Data

Column: $5C_{18}$-MS-II
Column size: 4.6mmI.D.-150mm
Mobile phase: Acetonitrile/ H_2O = 40/60
Flow rate: 1.0 ml/min
Temperature: 50°C
Detection: UV206nm

Sample: 1; Saikosaponin a (1.0µg)
2; Saikosaponin d (1.0µg)



NACALAI TESQUE, INC

AP-0910

(2) Crude Drugs

● Saireito Extract

COSMOSIL Application Data

Column: $5C_{18}$-MS-II
Column size: 4.6mmI.D.-150mm
Mobile phase: Acetonitrile/ 50mmol/l NaH_2PO_4 = 3/5
Flow rate: 1.0 ml/min
Temperature: 40°C
Detection: UV254nm

Sample: 1; Saikosaponin b_2 (1.0µg)

NACALAI TESQUE, INC

AP-0978

● Saireito Extract

COSMOSIL Application Data

Column: $5C_{18}$-MS-II
Column size: 4.6mmI.D.-150mm
Mobile phase: Acetonitrile/ 0.5%H_3PO_4 = 24/76
Flow rate: 1.0 ml/min
Temperature: 40°C
Detection: UV277nm

Sample: 1; Baicalin (0.5µg)

NACALAI TESQUE, INC

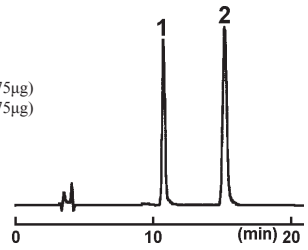
AP-0981

● Powdered Gardenia Fruit

COSMOSIL Application Data

Column: $5C_{18}$-MS-II
Column size: 4.6mmI.D.-150mm
Mobile phase: Acetonitrile/ H_2O = 60/440
Flow rate: 0.5 ml/min
Temperature: 30°C
Detection: UV240nm

Sample: 1; Caffeine (0.175µg)
2; Geniposide (0.175µg)



NACALAI TESQUE, INC

AP-0966

● Peony Root

COSMOSIL Application Data

Column: $5C_{18}$-AR-II
Column size: 4.6mmI.D.-150mm
Mobile phase: Acetonitrile/ 0.12%H_3PO_4 =15/85
Flow rate: 1.0 ml/min
Temperature: 30°C
Detection: UV232nm

Sample: 1; Albiflorin (1.0µg)
2; Paeoniflorin (1.0µg)

NACALAI TESQUE, INC

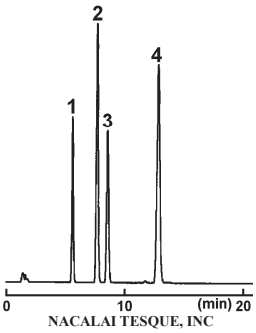
AP-0960

● Toad Venom

COSMOSIL Application Data

Column: $5C_{18}$-AR-II
Column size: 4.6mmI.D.-150mm
Mobile phase: Acetonitrile/ 0.1%H_3PO_4 = 45/55
Flow rate: 1.0 ml/min
Temperature: 40°C
Detection: UV300nm

Sample:
1; Bufalin (0.4µg)
2; Cinobufagin (0.4µg)
3; Resibufogenin (0.6µg)
4; Indometacin〔Indomethacin〕(0.6µg)



NACALAI TESQUE, INC

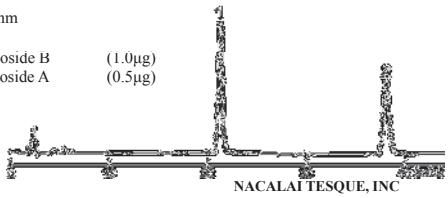
AP-1000

● Senna Leaf

COSMOSIL Application Data

Column: $5C_{18}$-MS-II
Column size: 4.6mmI.D.-250mm
Mobile phase: 5mmol/l Tetra-*n*-heptylammonium Bromide-Acetonitrile/ 0.1mol/l Acetic Acid, 1%Sodium Acetate(pH5.0) = 32/68
Flow rate: 1.0 ml/min
Temperature: 50°C
Detection: UV340nm

Sample: 1; Sennoside B (1.0µg)
2; Sennoside A (0.5µg)



NACALAI TESQUE, INC

AP-0993

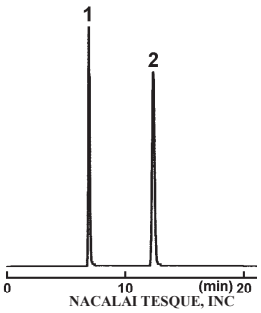
(2) Crude Drugs

● Swertia Herb

COSMOSIL Application Data

Column: $5C_{18}$-MS-II
Column size: 4.6mmI.D.-150mm
Mobile phase: Acetonitrile/ H_2O = 10/90
Flow rate: 0.5 ml/min
Temperature: 50°C
Detection: UV238nm

Sample: 1; Theophylline (1.0µg)
2; Swertiamarin (1.0µg)



NACALAI TESQUE, INC

AP-0996

● Rhubarb

COSMOSIL Application Data

Column: $5C_{18}$-MS-II
Column size: 4.6mmI.D.-150mm
Mobile phase: Acetonitrile/ 1.25%Acetic Acid = 20/80
Flow rate: 0.5 ml/min
Temperature: 40°C
Detection: UV340nm

Sample: 1; Sennoside A (2.0µg)
2; Naringin (2.0µg)

NACALAI TESQUE, INC

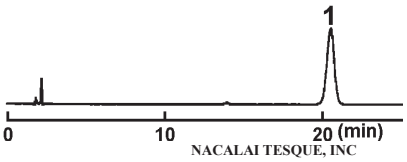
AP-0972

● Daiokanzoto Extract

COSMOSIL Application Data

Column: $5C_{18}$-PAQ
Column size: 4.6mmI.D.-150mm
Mobile phase: Acetonitrile/ 0.04%H_3PO_4 = 540/2460
Flow rate: 1.0 ml/min
Temperature: 30°C
Detection: UV340nm

Sample: 1; Sennoside A (0.26µg)



NACALAI TESQUE, INC

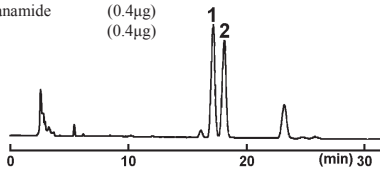
AP-0916

● Capsicum

COSMOSIL Application Data

Column: 5PE-MS
Column size: 4.6mmI.D.-250mm
Mobile phase: Acetonitrile/ 0.1%H_3PO_4 = 40/60
Flow rate: 1.0 ml/min
Temperature: 30°C
Detection: UV281nm

Sample: 1; *N*-Vanillylnonanamide (0.4µg)
2; Capsaicin (0.4µg)



NACALAI TESQUE, INC

AP-0913

● Ipecac

COSMOSIL Application Data

Column: $5C_{18}$-MS-II
Column size: 4.6mmI.D.-150mm
Mobile phase: Methanol/ 20mmol/l Sodium *1*-Heptanesulfonate (pH4.0 with Acetic Acid) = 50/50
Flow rate: 1.0 ml/min
Temperature: 50°C
Detection: UV283nm

Sample: 1; Cephaeline (2.0µg)
2; Emetine (0.5µg)

NACALAI TESQUE, INC

AP-0938

● Belladonna Root

COSMOSIL Application Data

Column: $5C_{18}$-PAQ
Column size: 4.6mmI.D.-150mm
Mobile phase: Acetonitrile/ 50mmol/l KH_2PO_4, 1%Triethylamine(pH3.5 with H_3PO_4) = 10/90
Flow rate: 1.0 ml/min
Temperature: 30°C
Detection: UV210nm

Sample: 1; Atropine (2.0µg)
2; Brucine (0.5µg)

NACALAI TESQUE, INC

AP-0909

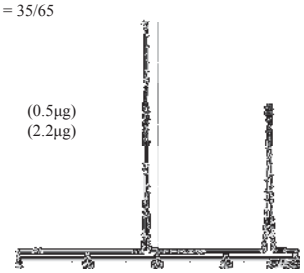
(2) Crude Drugs

● Moutan Bark

COSMOSIL Application Data

Column: $5C_{18}$-MS-II
Column size: 4.6mmI.D.-250mm
Mobile phase: Acetonitrile/ 3.1%Acetic Acid = 35/65
Flow rate: 1.0 ml/min
Temperature: 30°C
Detection: UV274nm

Sample: 1; Paeonol (0.5µg)
2; Butyl *p*-Hydroxybenzoate (2.2µg)



NACALAI TESQUE, INC

AP-0950

● Hochuekkito Extract

COSMOSIL Application Data

Column: $5C_{18}$-AR-II
Column size: 4.6mmI.D.-150mm
Mobile phase: Acetonitrile/ 1.2%H_3PO_4 = 18/82
Flow rate: 1.0 ml/min
Temperature: 40°C
Detection: UV285nm

Sample: 1; Naringin (0.1µg)
2; Vitamin P [Hesperidin] (0.1µg)

NACALAI TESQUE, INC

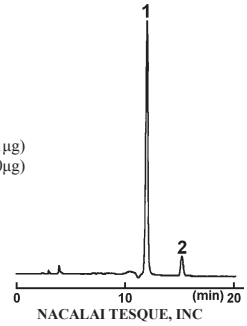
AP-0930

● Povidone

COSMOSIL Application Data

Column: $5C_8$-MS
Column size: 4.6mmI.D.-250mm
Mobile phase: Methanol/ H_2O = 20/80
Flow rate: 1.0 ml/min
Temperature: 40°C
Detection: UV254nm

Sample: 1; *N*-Vinyl-2-Pyrrolidone (0.1µg)
2; Vinyl Acetate (5.0µg)



NACALAI TESQUE, INC

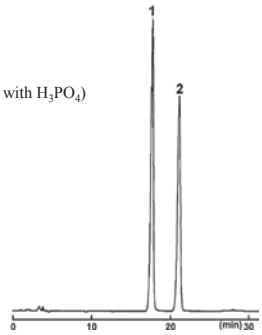
AP-0965

● Nux Vomica

COSMOSIL Application Data

Column: $5C_{18}$-PAQ
Column size: 4.6mmI.D.-150mm
Mobile phase: Acetonitrile/ 50mmol/l KH_2PO_4,
1.11%Triethylamine = 10/90(pH3.0 with H_3PO_4)
Flow rate: 0.5 ml/min
Temperature: 30°C
Detection: UV210nm

Sample: 1; Strychnine (0.8µg)
2; Barbital (1.1µg)



NACALAI TESQUE, INC

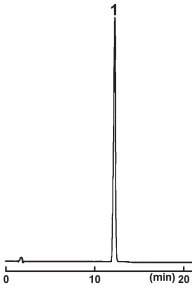
AP-0958

● Ryokeijutukanto Extract

COSMOSIL Application Data

Column: $5C_{18}$-AR-II
Column size: 4.6mmI.D.-150mm
Mobile phase: Acetonitrile/ 0.13%H_3PO_4 = 25/75
Flow rate: 1.0 ml/min
Temperature: 40°C
Detection: UV273nm

Sample: 1; *trans*-Cinnamic Acid (0.1µg)



NACALAI TESQUE, INC

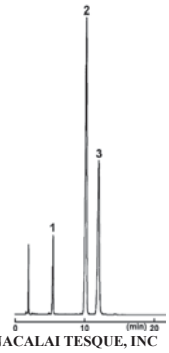
AP-0976

● Scopolia Rhizome and Scopolia Extract

COSMOSIL Application Data

Column: $5C_{18}$-PAQ
Column size: 4.6mmI.D.-150mm
Mobile phase: Acetonitrile/ 50mmol/l KH_2PO_4,
0.1%Triethylamine(pH3.5 with H_3PO_4)
= 10/90
Flow rate: 1.0 ml/min
Temperature: 30°C
Detection: UV210nm

Sample: 1; Scopolamine (0.6µg)
2; Atropine Sulfate (2.0µg)
3; Brucine (0.4µg)



NACALAI TESQUE, INC

AP-0989

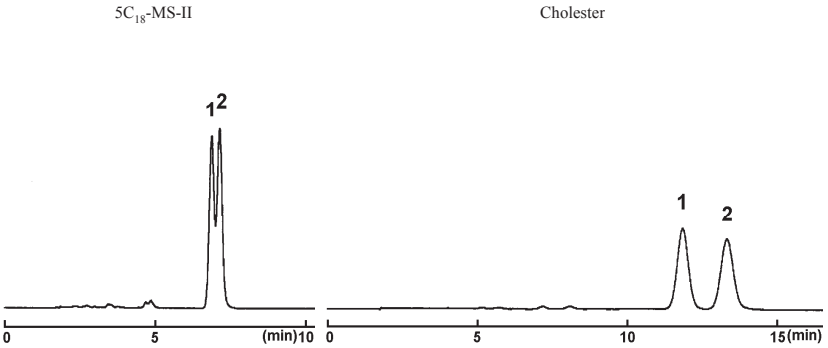
(3) Natural Compounds

● Carotenes

COSMOSIL Application Data

Column:
Column size: 4.6mmI.D.-150mm
Mobile phase: Tetrahydrofuran/Methanol = 20/80
Flow rate: 1.0 ml/min
Temperature: 30°C
Detection: UV470nm

Sample: 1; α-Carotene
2; β-Carotene



NACALAI TESQUE, INC

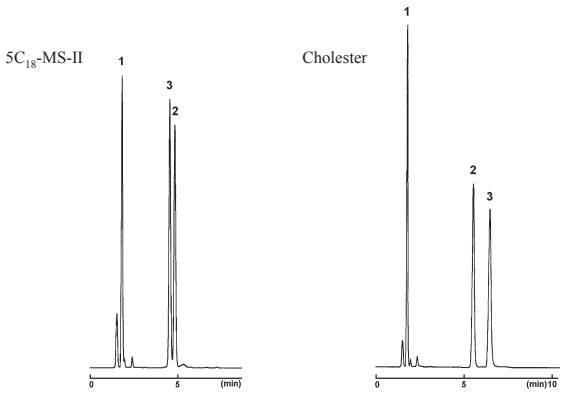
AP-1031

● Flavanones

COSMOSIL Application Data

Column:
Column size: 4.6mmI.D.-150mm
Mobile phase: Acetonitrile/ 20mmol/l Phosphate Buffer(pH2.5) = 40/60
Flow rate: 1.0 ml/min
Temperature: 30°C
Detection: UV280nm

Sample: 1; Naringin (0.4µg)
2; Naringenin (0.2µg)
3; Apigenin (0.2µg)



NACALAI TESQUE, INC

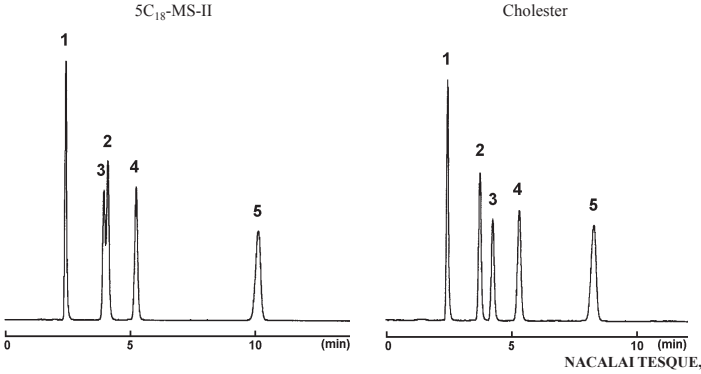
AP-1033

● Saikosaponins

COSMOSIL Application Data

Column:
Column size: 4.6mmI.D.-150mm
Mobile phase: Acetonitrile/ H_2O = 45/55
Flow rate: 1.0 ml/min
Temperature: 30°C
Detection: ELSD, Gain=6

Sample: 1; Saikosaponin c (1.5µg)
2; Saikosaponin a (1.5µg)
3; Saikosaponin b_2 (1.5µg)
4; Saikosaponin b_1 (1.5µg)
5; Saikosaponin d (1.5µg)



NACALAI TESQUE, INC

AP-1021

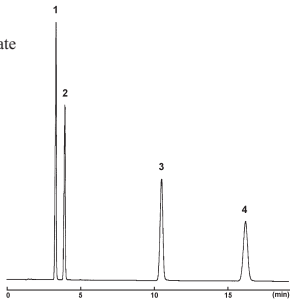
(3) Natural Compounds

● Hydroxyflavones

COSMOSIL Application Data

Column: $5C_{18}$-MS-II
Column size: 4.6mmI.D.-150mm
Mobile phase: Acetonitrile / 20mmol/l Phosphate Buffer(pH2.5) = 50/50
Flow rate: 1.0 ml/min
Temperature: 30°C
Detection: UV280nm

Sample:
1; *7*-Hydroxyflavone (0.2µg)
2; *6*-Hydroxyflavone (0.1µg)
3; *3*-Hydroxyflavone (0.5µg)
4; *5*-Hydroxyflavone (0.1µg)



NACALAI TESQUE, INC

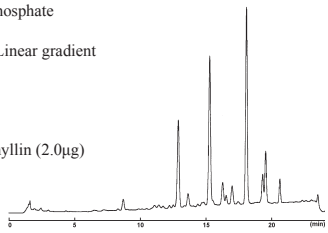
AP-0245

● Chlorophyll

COSMOSIL Application Data

Column: $5C_{18}$-MS-II
Column size: 4.6mmI.D.-150mm
Mobile phase: A: Methanol/ 20mmol/l Phosphate Buffer(pH7.0) = 60/40
B: Methanol/ 20mmol/l Phosphate Buffer(pH7.0) = 95/5
B conc. 0→100% 20min Linear gradient
Flow rate: 1.0 ml/min
Temperature: 30°C
Detection: UV405nm

Sample: Sodium Copper Chlorophyllin (2.0µg)



NACALAI TESQUE, INC

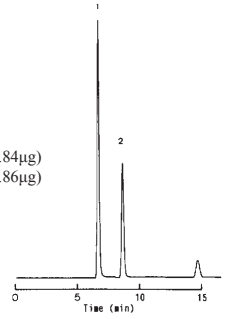
AP-0249

● Coumarins

COSMOSIL Application Data

Column: $5C_{18}$-MS-II
Column size: 4.6mmI.D.-150mm
Mobile phase: Methanol / H_2O = 40/60
Flow rate: 1.0 ml/min
Temperature: 30°C
Detection: UV254nm, 0.16AUFS

Sample:
1; Coumarin (0.84µg)
2; *3,4*-Dihydrocoumarin (3.86µg)



NACALAI TESQUE, INC

AP-0124

● Hinokitiol

COSMOSIL Application Data

Column: $5C_{18}$-AR-II
Column size: 4.6mmI.D.-150mm
Mobile phase: Methanol/ 1mmol/l EDTA, 20mmol/l Phophoric Acid = 70/30
Flow rate: 1.0 ml/min
Temperature: 30°C
Detection: UV254nm, 0.5AUFS

Sample: Hinokitiol (1.0µg)

NACALAI TESQUE, INC

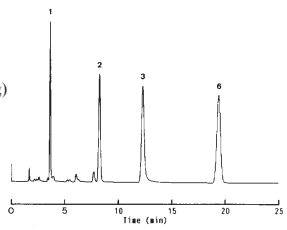
AP-0393

● Anthraquinone Dyes

COSMOSIL Application Data

Column: $5C_{18}$-MS-II
Column size: 4.6mmI.D.-150mm
Mobile phase: Methanol/ 20mmol/l Phosphate Buffer(pH3) = 75/25
Flow rate: 1.0 ml/min
Temperature: 30°C
Detection: UV254nm, 0.16AUFS

Sample:
1; Alizalin (0.05µg)
2; Chrysazin (0.1µg)
3; Anthrarufin (0.3µg)
6; Amylbenzene (10µg)



NACALAI TESQUE, INC

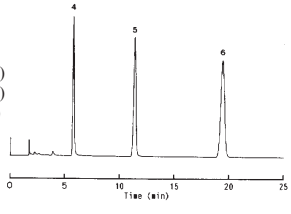
AP-0131

● Anthraquinone Dyes

COSMOSIL Application Data

Column: $5C_{18}$-MS-II
Column size: 4.6mmI.D.-150mm
Mobile phase: Methanol/ 20mmol/l Phosphate Buffer(pH3) = 75/25
Flow rate: 1.0 ml/min
Temperature: 30°C
Detection: UV254nm, 0.16AUFS

Sample:
4; Purpurin (0.2µg)
5; Quinizarin (0.1µg)
6; Amylbenzene (10µg)



NACALAI TESQUE, INC

AP-0132

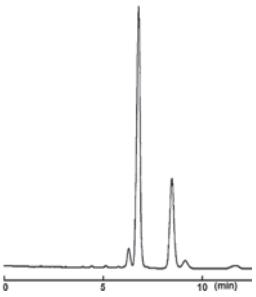
(3) Natural Compounds

● Capsaicin

COSMOSIL Application Data

Column: πNAP
Column size: 4.6mmI.D.-150mm
Mobile phase: Methanol/ H_2O = 70/30
Flow rate: 1.0 ml/min
Temperature: 30°C
Detection: UV280nm

Sample: Capsaicin (2.0µg)



NACALAI TESQUE, INC

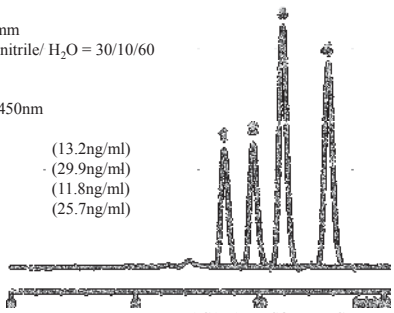
AP-0298

● Fungal Toxin

COSMOSIL Application Data

Column: $5C_{18}$-AR-II
Column size: 4.6mmI.D.-150mm
Mobile phase: Methanol/ Acetonitrile/ H_2O = 30/10/60
Flow rate: 0.8 ml/min
Temperature: 40°C
Detection: Ex.365nm Em.450nm

Sample: 1; Aflatoxin G2 (13.2ng/ml)
2; Aflatoxin G1 (29.9ng/ml)
3; Aflatoxin B2 (11.8ng/ml)
4; Aflatoxin B1 (25.7ng/ml)
Inj.Vol. 20µl



NACALAI TESQUE, INC

Data courtesy of a customer

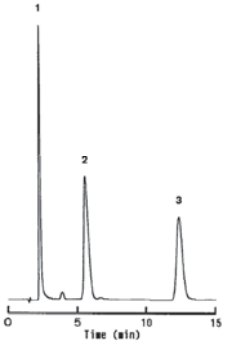
AP-1148

● Alkaloids

COSMOSIL Application Data

Column: $5C_{18}$-MS-II
Column size: 4.6mmI.D.-150mm
Mobile phase: Methanol/ 20mmol/l Phosphate buffer(pH3) = 50/50
Flow rate: 1.0 ml/min
Temperature: 30°C
Detection: UV220nm

Sample: 1; Papaverine (0.17µg)
2; Aconitine (8.1µg)
3; Reserpine (1.7µg)



NACALAI TESQUE, INC

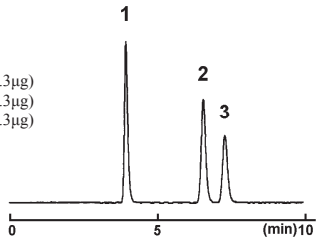
AP-0133

● Ginkgo Biloba

COSMOSIL Application Data

Column: $5C_{18}$-MS-II
Column size: 4.6mmI.D.-150mm
Mobile phase: Methanol/ H_2O = 40/60
Flow rate: 1.0 ml/min
Temperature: 30°C
Detection: ELSD

Sample: 1; Ginkgolide C (1.3µg)
2; Ginkgolide A (1.3µg)
3; Ginkgolide B (1.3µg)



NACALAI TESQUE, INC

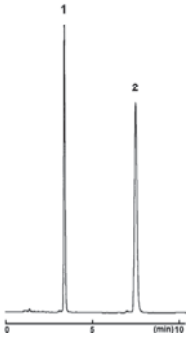
AP-1063

● Zingiberis Rhizoma

COSMOSIL Application Data

Column: $5C_{18}$-MS-II
Column size: 4.6mmI.D.-150mm
Mobile phase: Acetonitrile/ H_2O = 60/40
Flow rate: 1.0 ml/min
Temperature: 30°C
Detection: UV225nm

Sample: 1; *6*-Gingerol (0.31µg)
2; *6*-Shogaol (0.13µg)



NACALAI TESQUE, INC

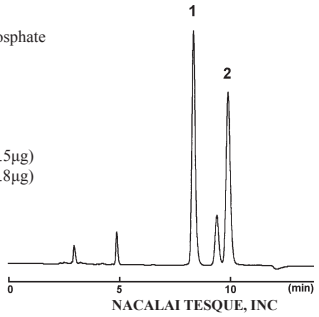
AP-1016

● Watermelon

COSMOSIL Application Data

Column: HILIC
Column size: 4.6mmI.D.-250mm
Mobile phase: Acetonitrile/ 20mmol/l Phosphate Buffer(pH7.0) = 70/30
Flow rate: 1.0 ml/min
Temperature: 30°C
Detection: UV210nm

Sample: 1; *L*-Citrulline (7.5µg)
2; *L*-(-)-Malic Acid (3.8µg)



NACALAI TESQUE, INC

AP-1062

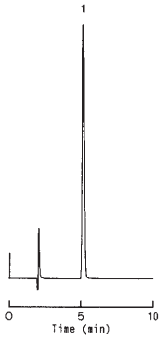
(4) Pesticides

● Asulam

COSMOSIL Application Data

Column: $5C_{18}$-MS-II
Column size: 4.6mmI.D.-150mm
Mobile phase: Acetonitrile/ 50mmol/l Phosphate Buffer(pH3) = 15/85
Flow rate: 1.0 ml/min
Temperature: 30°C
Detection: UV270nm

Sample: Asulam



NACALAI TESQUE, INC

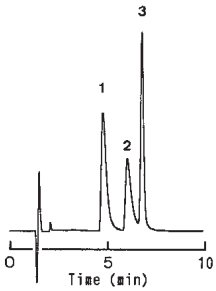
AP-0144

● Chlorophenoxyacetic Herbicides

COSMOSIL Application Data

Column: $5C_{18}$-MS-II
Column size: 4.6mmI.D.-150mm
Mobile phase: Acetonitrile/ 0.1%Acetic Acid = 50/50
Flow rate: 1.0 ml/min
Temperature: 30°C
Detection: UV275nm, 0.02AUFS

Sample: 1; MCP (0.22µg)
2; MCPP (0.20µg)
3; MCPB (0.24µg)



NACALAI TESQUE, INC

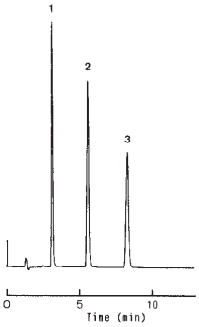
AP-0147

● Pesticides used at Golf Course

COSMOSIL Application Data

Column: $5C_{18}$-MS-II
Column size: 4.6mmI.D.-150mm
Mobile phase: Acetonitrile/ 50mmol/l Phosphate buffer(pH3) = 60/40
Flow rate: 1.0 ml/min
Temperature: 30°C
Detection: UV230nm

Sample: 1; Thiram
2; Iprodione
3; Bensulide



NACALAI TESQUE, INC

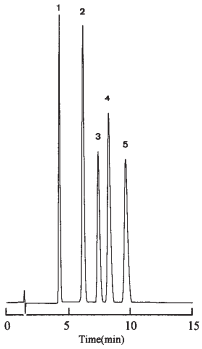
AP-0145

● Diphenyl Ether Herbicides

COSMOSIL Application Data

Column: $5C_{18}$-MS-II
Column size: 4.6mmI.D.-150mm
Mobile phase: Acetonitrile/ H_2O = 70/30
Flow rate: 1.0 ml/min
Temperature: 30°C
Detection: UV254nm, 0.1AUFS

Sample: 1; Fluorodifen (0.6µg)
2; Chlomethoxynil (1.0µg)
3; Nitrofen (1.0µg)
4; Oxyfluorfen (1.2µg)
5; CNP (1.0µg)



NACALAI TESQUE, INC

AP-0146

● Aniline Herbicides

COSMOSIL Application Data

Column: $5C_{18}$-MS-II
Column size: 4.6mmI.D.-150mm
Mobile phase: Acetonitrile/ H_2O = 50/50
Flow rate: 1.0 ml/min
Temperature: 30°C
Detection: UV254nm, 0.1AUFS

Sample: 1; DCMU (0.11µg)
2; DCPA (0.08µg)
3; Linuron (0.11µg)
4; MCC (0.18µg)

NACALAI TESQUE, INC

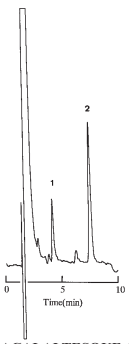
AP-0148

● Dithiocarbamate Herbicides

COSMOSIL Application Data

Column: $5C_{18}$-MS-II
Column size: 4.6mmI.D.-150mm
Mobile phase: Acetonitrile/ 20mmol/l KH_2PO_4 = 40/60
Flow rate: 1.0 ml/min
Temperature: 30°C
Detection: RI

Sample: 1; Maneb (60µg)
2; Thiram (10µg)



NACALAI TESQUE, INC

AP-0149

(4) Pesticides

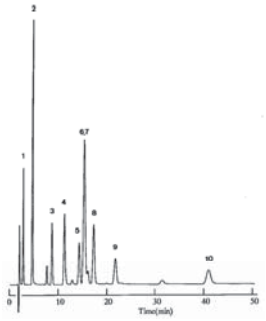
● Carbamate Herbicides

COSMOSIL Application Data

Column: $5C_{18}$-MS-II
Column size: 4.6mmI.D.-150mm
Mobile phase: Acetonitrile/ H_2O = 30/70
Flow rate: 1.0 ml/min
Temperature: 30°C
Detection: UV254nm, 0.05AUFS

Sample:

1;	Methomyl	(0.5μg)
2;	Pirimicarb	(0.5μg)
3;	MTMC	(2.0μg)
4;	PHC	(2.0μg)
5;	MPMC	(2.0μg)
6;	NAC	(1.0μg)
7;	XMC	(1.0μg)
8;	Ethiofencarb	(2.0μg)
9;	Isoprocarb	(2.0μg)
10;	BPMC	(2.0μg)



NACALAI TESQUE, INC

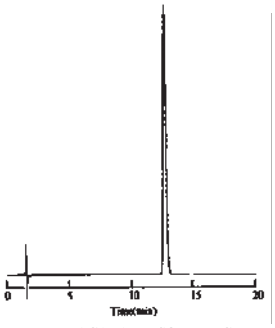
AP-0150

● Carbamate Herbicides

COSMOSIL Application Data

Column: $5C_{18}$-MS-II
Column size: 4.6mmI.D.-150mm
Mobile phase: Acetonitrile/ H_2O = 80/20
Flow rate: 1.0 ml/min
Temperature: 30°C
Detection: UV254nm, 0.05AUFS

Sample: Carbosulfan (6.0μg)



NACALAI TESQUE, INC

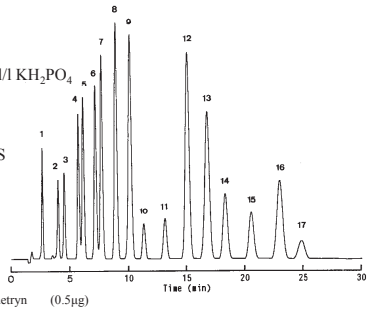
AP-0151

● Triazine and Urea Herbicides

COSMOSIL Application Data

Column: $5C_{18}$-MS-II
Column size: 4.6mmI.D.-150mm
Mobile phase: Acetonitrile/ 20mmol/l KH_2PO_4 = 40/60
Flow rate: 1.0 ml/min
Temperature: 30°C
Detection: UV254nm, 0.2AUFS
Sample:

1;	PAC	(0.5μg)	
2;	Bromacil	(0.5μg)	
3;	CAT	(0.5μg)	
4;	Methabenzthiazuron	(0.5μg)	
5;	Chlorotoluron	(0.5μg)	
6;	Isoproturon	(0.5μg)	
7;	Monolinuron	(0.5μg)	
8;	Metabromuron	(0.5μg)	14; Prometryn (0.5μg)
9;	Dimefuron	(0.5μg)	15; Terbutryn (0.5μg)
10;	Propazine	(0.5μg)	16; Chloro IPC (2.5μg)
11;	Terbuthylazine	(0.5μg)	17; Ethofumesate (5.0μg)
12;	Linuron	(0.5μg)	
13;	Chloroxuron	(0.5μg)	



NACALAI TESQUE, INC

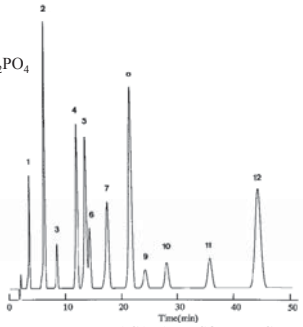
AP-0152

● Triazine and Urea Herbicides

COSMOSIL Application Data

Column: $5C_{18}$-MS-II
Column size: 4.6mmI.D.-150mm
Mobile phase: Acetonitrile/ 20mmol/l KH_2PO_4 = 30/70
Flow rate: 1.0 ml/min
Temperature: 30°C
Detection: UV254nm, 0.2AUFS
Sample:

1;	Ethidimuron	(1.0μg)
2;	Metoxuron	(1.0μg)
3;	Cyanazine	(1.0μg)
4;	Methabenzthiazuron	(1.0μg)
5;	Chlorotoluron	(1.0μg)
6;	Atrazine	(1.0μg)
7;	Isoproturon	(1.0μg)
8;	Metabromuron	(1.0μg)
9;	Metazachlor	(5.0μg)
10;	Propazine	(1.0μg)
11;	Terbuthylazine	(1.0μg)
12;	Linuron	(1.0μg)



NACALAI TESQUE, INC

AP-0153

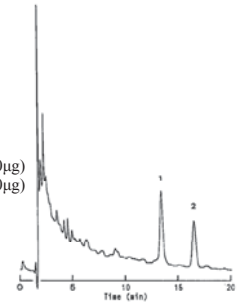
(5) Food Additives

● Natural Colorants (Chlorophyll)

COSMOSIL Application Data

Column: $5C_{18}$-AR-II
Column size: 4.6mmI.D.-150mm
Mobile phase: Methanol / 20mmol/l Phosphoric Acid = 30/70
Flow rate: 1.0 ml/min
Temperature: 30°C
Detection: UV254nm, 0.02AUFS

Sample: 1; Sodium Copper Chlorophyllin (30μg)
2; Sodium Iron Chlorophyllin (30μg)



NACALAI TESQUE, INC

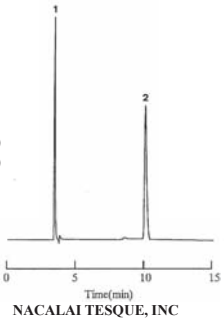
AP-0095

● Natural Colorants (Carotenoid)

COSMOSIL Application Data

Column: 5SL-II
Column size: 4.6mmI.D.-250mm
Mobile phase: Chloroform/Hexane = 1/9
Flow rate: 1.0 ml/min
Temperature: 30°C
Detection: UV254nm, 0.16AUFS

Sample: 1; β-Carotene (0.5μg)
2; Vitamin A Acetate, *all trans* (1.5μg)



NACALAI TESQUE, INC

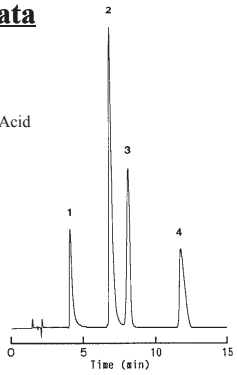
AP-0094

● Synthetic Sweeteners

COSMOSIL Application Data

Column: $5C_{18}$-MS-II
Column size: 4.6mmI.D.-150mm
Mobile phase: Acetonitrile / 20mmol/l Phosphoric Acid = 10/90
Flow rate: 1.0 ml/min
Temperature: 30°C
Detection: UV210nm, 1.0AUFS

Sample: 1; Acesulfame (1.0μg)
2; Saccharin (1.0μg)
3; Diketopiperazine (1.0μg)
4; Aspartame (1.0μg)



NACALAI TESQUE, INC

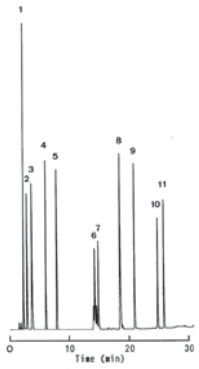
AP-0107

● Synthetic Colorants

COSMOSIL Application Data

Column: $5C_{18}$-MS-II
Column size: 4.6mmI.D.-150mm
Mobile phase: A; Methanol/ 20mmol Phosphate buffer(pH7) = 10/90
B; Methanol/ 20mmol Phosphate buffer(pH7) = 80/20
B conc. 15→100% Linear gradient
Flow rate: 1.0 ml/min
Temperature: 30°C
Detection: UV254nm, 0.64AUFS
Sample:

1; Tartrazine	(1.0μg)	7; Brilliant Blue FCF	(3.0μg)
2; Amaranth	(1.0μg)	8; Acid Red 52	(1.0μg)
3; Indigo Carmine	(1.0μg)	9; Erythrosine B	(1.0μg)
4; New Coccine	(1.0μg)	10; Phloxine B	(1.0μg)
5; Sunset Yellow FCF	(1.0μg)	11; Rose Bengal	(1.0μg)
6; Fast Green FCF	(3.0μg)		



NACALAI TESQUE, INC

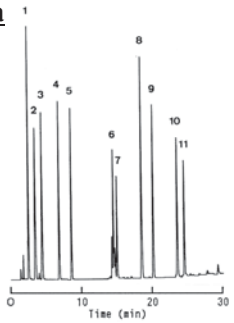
AP-0092

● Synthetic Colorants

COSMOSIL Application Data

Column: $5C_{18}$-AR-II
Column size: 4.6mmI.D.-150mm
Mobile phase: A; Methanol/ 20mmol Phosphate buffer(pH7) = 10/90
B; Methanol/ 20mmol Phosphate buffer(pH7) = 80/20
B conc. 15→100% Linear gradient
Flow rate: 1.0 ml/min
Temperature: 30°C
Detection: UV254nm, 0.64AUFS
Sample:

1; Tartrazine	(1.0μg)	7; Brilliant Blue FCF	(3.0μg)
2; Amaranth	(1.0μg)	8; Acid Red 52	(1.0μg)
3; Indigo Carmine	(1.0μg)	9; Erythrosine B	(1.0μg)
4; New Coccine	(1.0μg)	10; Phloxine B	(1.0μg)
5; Sunset Yellow FCF	(1.0μg)	11; Rose Bengal	(1.0μg)
6; Fast Green FCF	(3.0μg)		



NACALAI TESQUE, INC

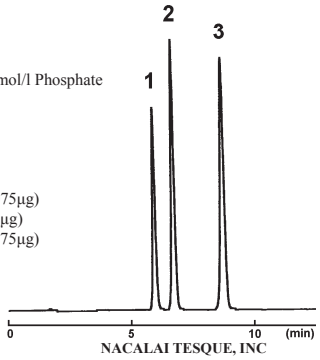
AP-0093

● Food Preservatives

COSMOSIL Application Data

Column: $5C_{18}$-MS-II
Column size: 4.6mmI.D.-150mm
Mobile phase: Acetonitrile/Methanol/ 20mmol/l Phosphate Buffer(pH4.0) = 20/10/70
Flow rate: 1.0 ml/min
Temperature: 30°C
Detection: UV230nm

Sample: 1; Benzoic Acid (0.075μg)
2; Sorbic Acid (0.1μg)
3; Dehydroacetic Acid (0.175μg)



NACALAI TESQUE, INC

AP-0378

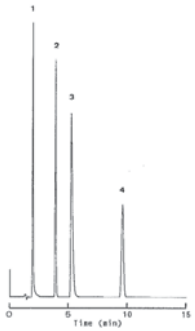
(5) Food Additives

● Preservatives (Fungicides)

COSMOSIL Application Data

Column: $5C_{18}$-MS-II
Column size: 4.6mmI.D.-150mm
Mobile phase: Acetonitrile/ H_2O = 60/40
Flow rate: 1.0 ml/min
Temperature: 30°C
Detection: UV230nm, 0.5AUFS

Sample:
1; *2*-(*4*-Thiazolyl)benzimidazole (0.3μg)
2; *o*-Phenylphenol (0.3μg)
3; Imazalil (2.1μg)
4; Biphenyl (0.3μg)



NACALAI TESQUE, INC

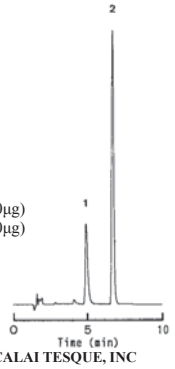
AP-0097

● Glycyrrhizic Acid

COSMOSIL Application Data

Column: $5C_{18}$-AR-II
Column size: 4.6mmI.D.-150mm
Mobile phase: Acetonitrile / (Acetic Acid/H_2O=1/15) = 2/3
Flow rate: 1.0 ml/min
Temperature: 30°C
Detection: UV254nm, 0.64AUFS

Sample: 1; Glycyrrhizic Acid〔Glycyrrhizin〕 (5.0μg)
2; *n*-Propyl *p*-Hydroxybenzoate (1.0μg)



NACALAI TESQUE, INC

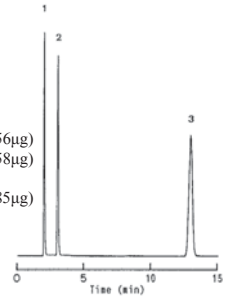
AP-0109

● Antioxidants

COSMOSIL Application Data

Column: $5C_{18}$-MS-II
Column size: 4.6mmI.D.-150mm
Mobile phase: Methanol / H_2O = 80/20
Flow rate: 1.0 ml/min
Temperature: 30°C
Detection: UV280nm, 0.16AUFS

Sample: 1; *tert*-Butylhydroquinone (0.56μg)
2; *3-tert*-Butyl-*4*-hydroxyanisol (0.58μg)
3; *2,6*-Di-*t*-butyl-*p*-cresol 〔Butylated Hydroxytoluene;BHT〕(1.85μg)



NACALAI TESQUE, INC

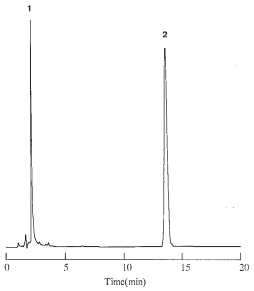
AP-0100

● Antioxidants

COSMOSIL Application Data

Column: $5C_{18}$-MS-II
Column size: 4.6mmI.D.-150mm
Mobile phase: Methanol / H_2O = 90/10
Flow rate: 1.0 ml/min
Temperature: 30°C
Detection: UV254nm, 0.32AUFS

Sample: 1; *n*-Propyl Gallate (1.0μg)
2; *6*-Ethoxy-*2,2,4*-Trimethyl -*1,2*-Dihydroquinoline (1.0μg)



NACALAI TESQUE, INC

AP-0101

● Repellents

COSMOSIL Application Data

Column: $5C_{18}$-AR-II
Column size: 4.6mmI.D.-150mm
Mobile phase: Methanol / H_2O = 90/10
Flow rate: 1.0 ml/min
Temperature: 30°C
Detection: UV254nm, 0.32AUFS

Sample: Piperonyl Butoxide (10μg)

NACALAI TESQUE, INC

AP-0104

● Umami Seasonings (Nucleic Acids)

COSMOSIL Application Data

Column: $5C_{18}$-AR-II
Column size: 4.6mmI.D.-150mm
Mobile phase: Acetonitrile/ 5mmol/l Tributylammonium bromide, 20mmol/l Phosphate buffer(pH7) = 5/95
Flow rate: 1.0 ml/min
Temperature: 30°C
Detection: UV270nm, 0.32AUFS

Sample: 1; Cytidine-*5′*-monophosphate (1.0μg)
2; Uridine-*5′*-monophosphate (1.0μg)
3; Guanosine-*5′*-monophosphate (1.0μg)
4; Inosine-*5′*-monophosphate (1.0μg)

NACALAI TESQUE, INC

AP-0106

(6) Vitamins

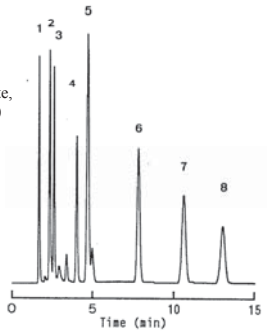
● Hydrosoluble Vitamins

COSMOSIL Application Data

Column: $5C_{18}$-MS-II
Column size: 4.6mmI.D.-150mm
Mobile phase: Acetonitrile / 5mmol/l Sodium *1*-Hexanesulfonate, 20mmol/l Phosphoric Acid = 10/90
Flow rate: 1.0 ml/min
Temperature: 30°C
Detection: UV210nm, 0.16AUFS

Sample:

1; *L*(+)-Ascorbic Acid [Vitamin C]	(0.23µg)
2; Nicotinic Acid	(0.057µg)
3; Nicotinamide	(0.042µg)
4; Vitamin B_6 [Pyridoxine]	(0.040µg)
5; Flavin Mononucleotide [FMN]	(0.19µg)
6; Vitamin B_1 [Thiamine]	(0.19µg)
7; Folic Acid	(0.084µg)
8; Vitamin B_2 [Riboflavin]	(0.57µg)



NACALAI TESQUE, INC

AP-0059

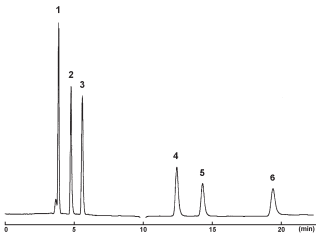
● Hydrosoluble Vitamins

COSMOSIL Application Data

Column: HILIC
Column size: 4.6mmI.D.-250mm
Mobile phase: Acetonitrile/ 100mmol/l Ammonium Acetate = 80/20
Flow rate: 1.0 ml/min
Temperature: 30°C
Detection: UV220nm

Sample:

1; Nicotinamide	(0.125µg)
2; Vitamin B_6 [Pyridoxine]	(0.25µg)
3; Vitamin B_2 [Riboflavin]	(0.25µg)
4; Nicotinic Acid	(0.125µg)
5; *D*-Pantothenic Acid	(3.125µg)
6; *L*(+)-Ascorbic Acid [Vitamin C]	(0.875µg)



NACALAI TESQUE, INC

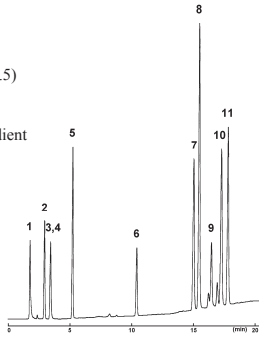
AP-0315

● Water-soluble Vitamins

COSMOSIL Application Data

Column: $5C_{18}$-MS-II
Column size: 4.6mmI.D.-150mm
Mobile phase: A; 20mmol/l Phosphate buffer(pH2.5)
B; Methanol/ 20mmol/l Phosphate Buffer(pH2.5) = 60/40
B conc. 0→80% 20min Linear gradient
Flow rate: 1.0 ml/min
Temperature: 30°C
Detection: UV220nm
Sample:

1; Vitamin B_1 [Thiamine]	(0.17µg)
2; *L*(+)-Ascorbic Acid [Vitamin C]	(0.33µg)
3; Nicotinic Acid	(0.03µg)
4; Nicotinamide	(0.05µg)
5; Vitamin B_6 [Pyridoxine]	(0.27µg)
6; *D*-Pantothenic Acid	(2.01µg)
7; Vitamin B_{12} [Cyanocobalamin]	(0.20µg)
8; Folic Acid	(0.26µg)
9; *D*-Biotin [Vitamin H]	(2.02µg)
10; Flavin Mononucleotide [FMN]	(0.26µg)
11; Vitamin B_2 [Riboflavin]	(0.13µg)



NACALAI TESQUE, INC

AP-1055

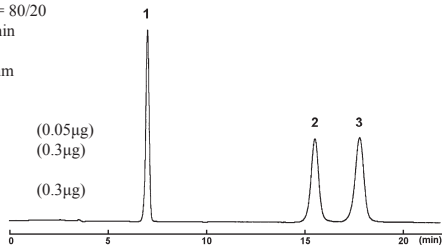
● Ascorbic Acids

COSMOSIL Application Data

Column: HILIC
Column size: 4.6mmI.D.-250mm
Mobile phase: Acetonitrile/ 100mmol/l Ammonium Acetate = 80/20
Flow rate: 1.0 ml/min
Temperature: 30°C
Detection: UV254nm

Sample:

1; Sorbic Acid	(0.05µg)
2; *D*-Isoascorbic Acid	(0.3µg)
3; *L*(+)-Ascorbic Acid [Vitamin C]	(0.3µg)



NACALAI TESQUE, INC

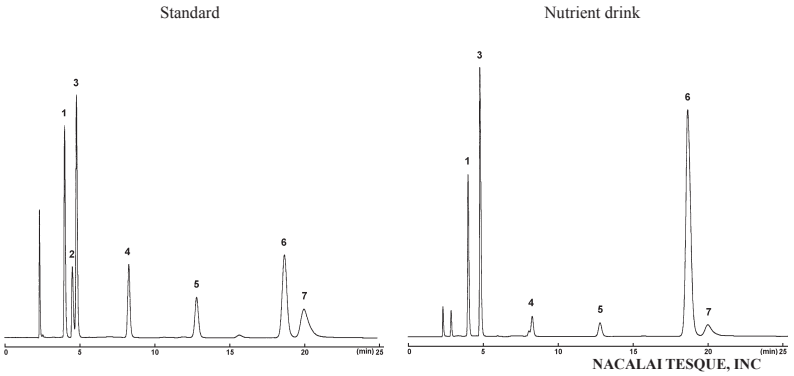
AP-0318

● Energy Drink

COSMOSIL Application Data

Column: $5C_{18}$-PAQ
Column size: 4.6mmI.D.-250mm
Mobile phase: Methanol/ 5mmol/l Sodium *1*-Hexanesulfonate, 20mmol/l Phosphoric Acid = 15/85
Flow rate: 1.0 ml/min
Temperature: 30°C
Detection: UV220nm

		Standard
Sample:	1; Citric Acid	(10mg/ml)
	2; *L*-Carnitine	(20mg/ml)
	3; Nicotinamide	(0.2mg/ml)
	4; Vitamin B_6 [Pyridoxine]	(0.2mg/ml)
	5; Vitamin B_1 [Thiamine]	(0.2mg/ml)
	6; Caffeine	(0.2mg/ml)
	7; Flavin Mononucleotide [FMN]	(0.2mg/ml)
Injection Vol.	1.0µl	



NACALAI TESQUE, INC

AP-1048

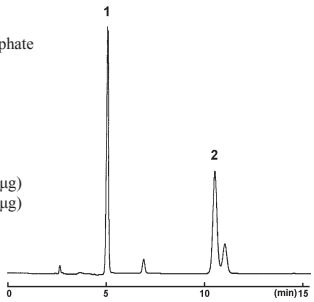
(6) Vitamins

● Fruit Juice

COSMOSIL Application Data

Column: HILIC
Column size: 4.6mmI.D.-250mm
Mobile phase: Acetonitrile/ 10mmol/l Phosphate Buffer(pH7.0) = 50/50
Flow rate: 1.0 ml/min
Temperature: 30°C
Detection: UV210nm

Sample:
1; *L*(+)-Ascorbic Acid [Vitamin C] (1.5µg)
2; *L*-(-)-Malic Acid (3.0µg)



NACALAI TESQUE, INC

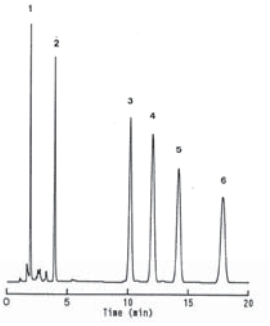
AP-0313

● Fat-soluble Vitamins

COSMOSIL Application Data

Column: $5C_{18}$-MS-II
Column size: 4.6mmI.D.-150mm
Mobile phase: Acetonitrile/Methanol = 60/40
Flow rate: 1.0 ml/min
Temperature: 30°C
Detection: UV280nm, 0.64AUFS

Sample:
1; Vitamin K_3 (1.71µg)
2; Vitamin A Acetate, *all trans* (0.44µg)
3; Vitamin D_3 (0.57µg)
4; Vitamin E [*DL*-α-Tocopherol] (4.97µg)
5; Vitamin E Acetate [*DL*-α-Tocopherol Acetate] (4.79µg)
6; Vitamin K_1 (1.68µg)



NACALAI TESQUE, INC

AP-0060

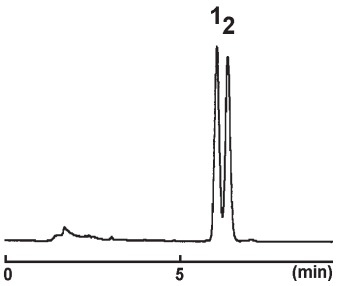
● Vitamin D

COSMOSIL Application Data

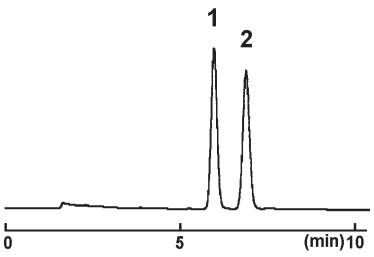
Column:
Column size: 4.6mmI.D.-150mm
Mobile phase: Methanol
Flow rate: 1.0 ml/min
Temperature: 30°C
Detection: UV265nm

Sample: 1; Vitamin D_2 [Calciferol] (0.3µg)
2; Vitamin D_3 (0.1µg)

$5C_{18}$-MS-II



Cholester



NACALAI TESQUE, INC

AP-1035

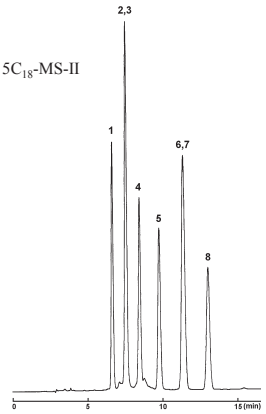
● Vitamin E

COSMOSIL Application Data

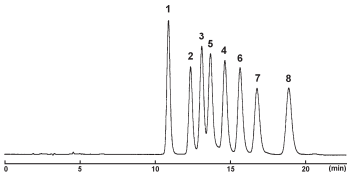
Column:
Column size: 4.6mmI.D.-250mm
Mobile phase: $5C_{18}$-MS-II Methanol
πNAP Methanol/ H_2O = 90/10
Flow rate: 1.0 ml/min
Temperature: 30°C
Detection: UV295nm

Sample:
1; δ-Tocotrienol
2; γ-Tocotrienol
3; β-Tocotrienol
4; α-Tocotrienol
5; δ-Tocopherol
6; γ-Tocopherol
7; β-Tocopherol
8; α-Tocopherol

$5C_{18}$-MS-II



πNAP



NACALAI TESQUE, INC

AP-1071

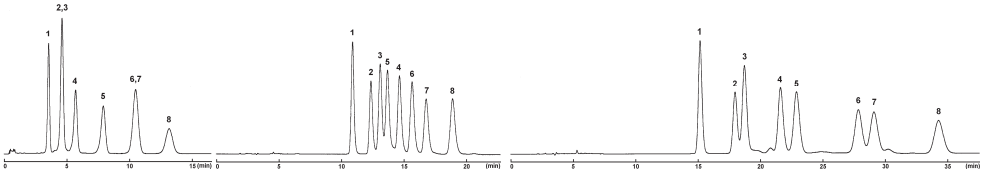
(6) Vitamins

● Vitamin E

COSMOSIL Application Data

	$5C_{18}$-MS-II (4.6mmI.D.-30mm)	πNAP (4.6mmI.D.-250mm)	πNAP (4.6mmI.D.-250mm) + $5C_{18}$-MS-II (4.6mmI.D.-30mm)
Column:			
Column size:			
Mobile phase:	Methanol/ H_2O = 90/10		
Flow rate:	1.0 ml/min		
Temperature:	30°C		
Detection:	UV295nm		

Sample:
1; δ-Tocotrienol
2; γ-Tocotrienol
3; β-Tocotrienol
4; α-Tocotrienol
5; δ-Tocopherol
6; γ-Tocopherol
7; β-Tocopherol
8; α-Tocopherol



NACALAI TESQUE, INC

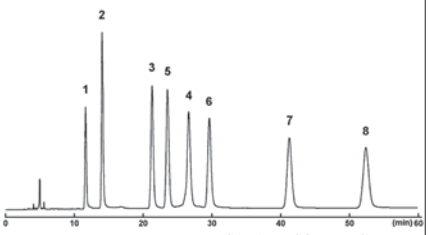
AP-1156

● Vitamin E

COSMOSIL Application Data

Column: SL-II
Column size: 4.6mmI.D.-250mm
Mobile phase: Hexane/ Acetic Acid = 99/1
Flow rate: 1.0 ml/min
Temperature: 30°C
Detection: UV295nm

Sample:
1; α-Tocopherol
2; β-Tocopherol
3; γ-Tocopherol
4; δ-Tocopherol
5; α-Tocotrienol
6; β-Tocotrienol
7; γ-Tocotrienol
8; δ-Tocotrienol



NACALAI TESQUE, INC

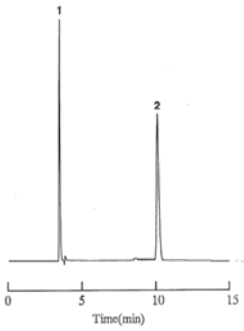
AP-1191

● Vitamin A

COSMOSIL Application Data

Column: 5SL-II
Column size: 4.6mmI.D.-250mm
Mobile phase: *n*-Hexane/Chloroform = 9/1
Flow rate: 1.0 ml/min
Temperature: 30°C
Detection: UV254nm, 0.16AUFS

Sample:
1; β-Carotene (0.5µg)
2; Vitamin A (1.5µg)



NACALAI TESQUE, INC

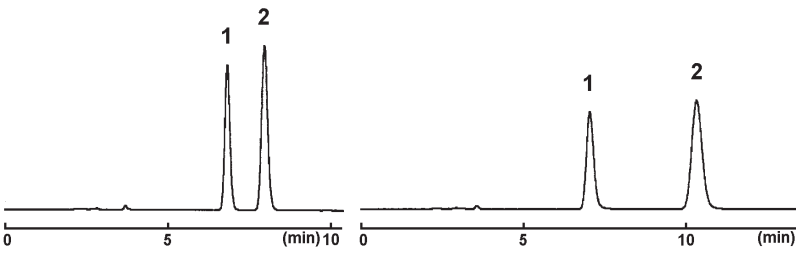
AP-0062

● Vitamin A Acid

COSMOSIL Application Data

	$5C_{18}$-MS-II	Cholester
Column:		
Column size:	4.6mmI.D.-150mm	
Mobile phase:	Methanol / 20mmol/l Phosphate Buffer(pH2.5) = 90/10	
Flow rate:	1.0 ml/min	
Temperature:	30°C	
Detection:	UV350nm	

Sample:
1; *13-cis*-Retinoic Acid (0.04µg)
2; Vitamin A Acid, *all-trans* 〔*all-trans*-Retinoic Acid〕 (0.04µg)



NACALAI TESQUE, INC

AP-1036

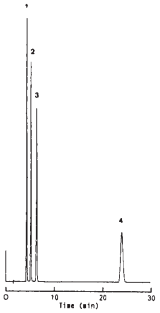
(7) Metabolites

● Androgens

COSMOSIL Application Data

Column: $5C_{18}$-AR-II
Column size: 4.6mmI.D.-150mm
Mobile phase: Methanol / H_2O = 70/30
Flow rate: 1.0 ml/min
Temperature: 30°C
Detection: UV235nm, 0.5AUFS

Sample:
1; *4*-Androstene-*3,17*-dione (1.0µg)
2; Testosterone (1.0µg)
3; *17*-Methyltestosterone (1.0µg)
4; Testosterone Propionate (1.0µg)



NACALAI TESQUE, INC

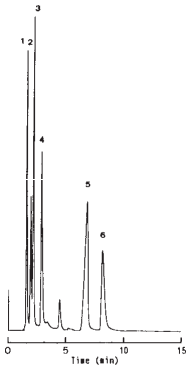
AP-0111

● Catecholamines

COSMOSIL Application Data

Column: $5C_{18}$-AR-II
Column size: 4.6mmI.D.-150mm
Mobile phase: 20mmol/l NaH_2PO_4
Flow rate: 1.0 ml/min
Temperature: 30°C
Detection: UV270nm, 0.1AUFS

Sample:
1; *L*-Noradrenaline (1.0µg)
2; (±)-Epinephrine (1.2µg)
3; *L*-DOPA (1.2µg)
4; Dopamine (1.0µg)
5; *3,4*-Dihydroxyphenylacetic Acid (1.0µg)
6; *3*-Methoxytyramine (1.0µg)



NACALAI TESQUE, INC

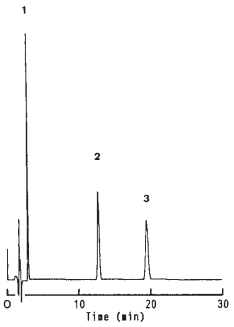
AP-0120

● Estrogens

COSMOSIL Application Data

Column: $5C_{18}$-AR-II
Column size: 4.6mmI.D.-150mm
Mobile phase: Acetonitrile/ H_2O = 35/65
Flow rate: 1.0 ml/min
Temperature: 30°C
Detection: UV280nm, 0.2AUFS

Sample:
1; Estriol (1.5µg)
2; *17*β-Estradiol (1.5µg)
3; Estrone (1.5µg)



NACALAI TESQUE, INC

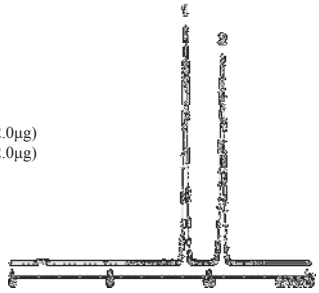
AP-0114

● Estradiols

COSMOSIL Application Data

Column: $5C_{18}$-MS-II
Column size: 4.6mmI.D.-150mm
Mobile phase: Acetonitrile/ H_2O = 40/60
Flow rate: 1.0 ml/min
Temperature: 30°C
Detection: UV280nm

Sample:
1; *17*β-Estradiol (2.0µg)
2; *17*α-Estradiol (2.0µg)



NACALAI TESQUE, INC

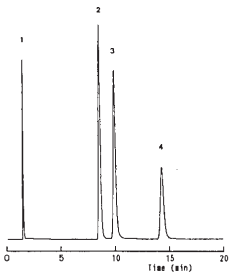
AP-0243

● Hippuric Acids

COSMOSIL Application Data

Column: $5C_{18}$-MS-II
Column size: 4.6mmI.D.-150mm
Mobile phase: Methanol/ 20mmol/l Phosphate Buffer(pH3) = 15/85
Flow rate: 1.0 ml/min
Temperature: 30°C
Detection: UV225nm, 0.32AUFS

Sample:
1; Creatinine (0.14µg)
2; Mandelic Acid (1.89µg)
3; Hippuric Acid (0.61µg)
4; *N*-(*o*-Toluoyl)glycine (0.52µg)



NACALAI TESQUE, INC

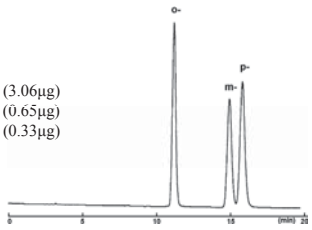
AP-0112

● Methylhippuric Acids

COSMOSIL Application Data

Column: 5PYE
Column size: 4.6mmI.D.-250mm
Mobile phase: Methanol/ 20mmol/l Phosphate buffer(pH2.5) = 40/60
Flow rate: 1.0 ml/min
Temperature: 30°C
Detection: UV254nm

Sample:
N-(*o*-Toluoyl)glycine (3.06µg)
N-(*m*-Toluoyl)glycine (0.65µg)
N-(*p*-Toluoyl)glycine (0.33µg)



NACALAI TESQUE, INC

AP-0289

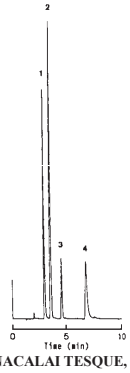
(7) Metabolites

● Urate Metabolites

COSMOSIL Application Data

Column: $5C_{18}$-AR-II
Column size: 4.6mmI.D.-150mm
Mobile phase: 20mmol/l Phosphoric Acid
Flow rate: 1.0 ml/min
Temperature: 30°C
Detection: UV280nm, 0.1AUFS

Sample:
1; Hypoxanthine (0.9µg)
2; Uric Acid (18.3µg)
3; Xanthine (9.0µg)
4; Allopurinol (1.8µg)



NACALAI TESQUE, INC

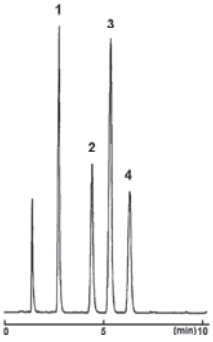
AP-0117

● Prostaglandins

COSMOSIL Application Data

Column: $5C_{18}$-MS-II
Column size: 4.6mmI.D.-150mm
Mobile phase: 0.05%TFA-40%Acetonitrile
Flow rate: 1.0 ml/min
Temperature: 30°C
Detection: ELSD

Sample:
1; Prostaglandin I_2 (2.0µg)
2; Prostaglandin $F_2\alpha$ (2.0µg)
3; Prostaglandin E_2 (2.0µg)
4; Prostaglandin D_2 (2.0µg)



NACALAI TESQUE, INC

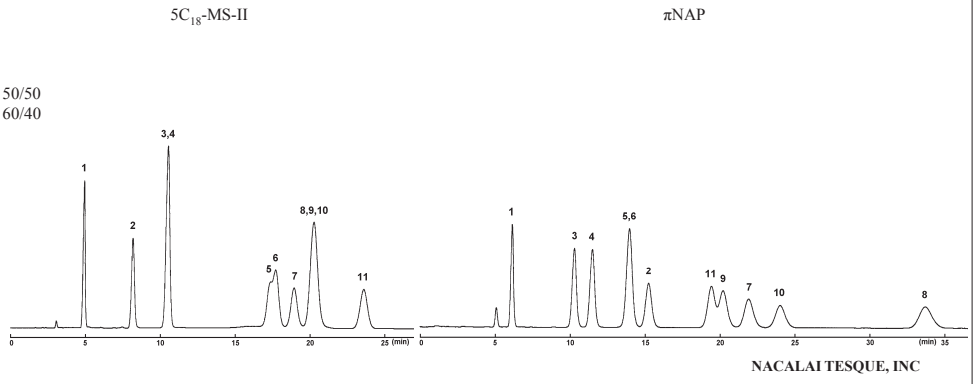
AP-0247

● Adrenal Cortical Hormones

COSMOSIL Application Data

Column:
Column size: 4.6mmI.D.-150mm
Mobile phase: $5C_{18}$-MS-II Methanol/ H_2O = 50/50
πNAP Methanol/ H_2O = 60/40
Flow rate: 1.0 ml/min
Temperature: 30°C
Detection: UV254nm

Sample:
1; Triamcinolone
2; Cortisone
3; Prednisolone
4; Hydrocortisone
5; Betamethasone
6; Dexamethasone
7; Triamcinolone Acetonide
8; Cortisone-*21*-Acetate
9; Prednisolone *21*-Acetate
10; Fluocinolone Acetonide
11; Fluorometholone
(each 0.23µg)



NACALAI TESQUE, INC

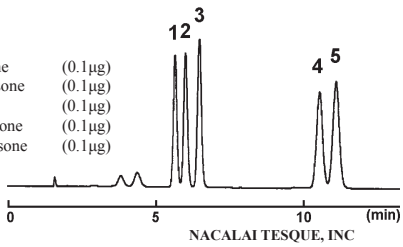
AP-1037

● Adrenal Cortical Hormones

COSMOSIL Application Data

Column: $5C_{18}$-MS-II
Column size: 4.6mmI.D.-150mm
Mobile phase: Acetonitrile/ H_2O = 30/70
Flow rate: 1.0 ml/min
Temperature: 30°C
Detection: UV240nm

Sample:
1; Prednisolone (0.1µg)
2; Hydrocortisone (0.1µg)
3; Cortisone (0.1µg)
4; Betamethasone (0.1µg)
5; Dexamethasone (0.1µg)



NACALAI TESQUE, INC

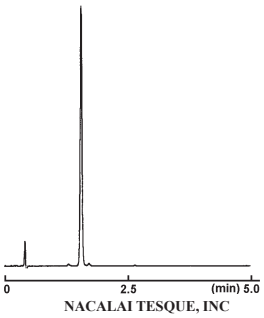
AP-0233

● Adrenal Cortical Hormones

COSMOSIL Application Data

Column: $2.5C_{18}$-MS-II
Column size: 3.0mmI.D.-75mm
Mobile phase: Acetonitrile/ H_2O = 30/70
Flow rate: 1.0 ml/min
Temperature: 40°C
Detection: UV242nm

Sample: Prednisone (0.02mg/ml)
Inj.Vol. 5µl



NACALAI TESQUE, INC

AP-1145

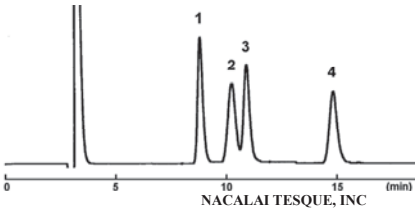
(8) Carbohydrates

● Oligosaccharides

COSMOSIL Application Data

Column: Sugar-D
Column size: 4.6mmI.D.-250mm
Mobile phase: Acetonitrile / H_2O = 75/25
Flow rate: 1.0 ml/min
Temperature: 30°C
Detection: RI

Sample:
1; Sucrose (10µg)
2; Maltose (10µg)
3; *D*-(+)- Trehalose (10µg)
4; *D*-(+)- Raffinose (10µg)



NACALAI TESQUE, INC

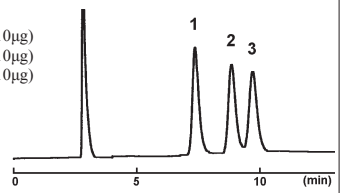
AP-0335

● Cyclodextrins

COSMOSIL Application Data

Column: Sugar-D
Column size: 4.6mmI.D.-250mm
Mobile phase: Acetonitrile / H_2O = 65/35
Flow rate: 1.0 ml/min
Temperature: 30°C
Detection: RI

Sample: 1; α-Cyclodextrin (10µg)
2; β-Cyclodextrin (10µg)
3; γ-Cyclodextrin (10µg)



NACALAI TESQUE, INC

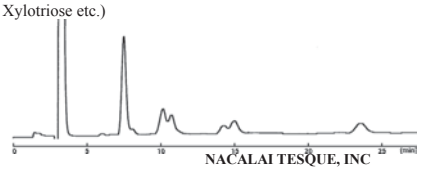
AP-0336

● Xylooligosaccharides

COSMOSIL Application Data

Column: Sugar-D
Column size: 4.6mmI.D.-250mm
Mobile phase: Acetonitrile / H_2O = 75/25
Flow rate: 1.0 ml/min
Temperature: 30°C
Detection: RI

Sample: Xylooligosaccharides (50µg)
(Xylobiose, Xylotriose etc.)



NACALAI TESQUE, INC

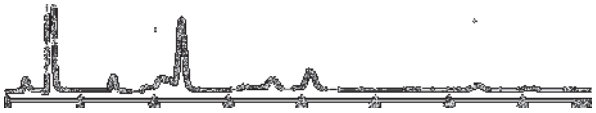
AP-0321

● Isomaltooligosaccharides

COSMOSIL Application Data

Column: Sugar-D
Column size: 4.6mmI.D.-250mm
Mobile phase: Acetonitrile / H_2O = 75/25
Flow rate: 1.0 ml/min
Temperature: 30°C
Detection: RI

Sample: Isomaltooligosaccharides (50µg)
(Isomaltose, Isomaltotriose, Panose etc.)



NACALAI TESQUE, INC

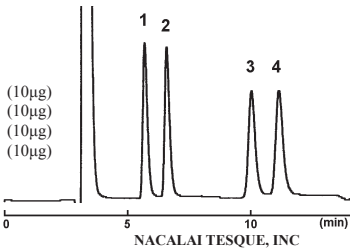
AP-0322

● Anticarious Foods Components

COSMOSIL Application Data

Column: Sugar-D
Column size: 4.6mmI.D.-250mm
Mobile phase: Acetonitrile / H_2O = 75/25
Flow rate: 1.0 ml/min
Temperature: 30°C
Detection: RI

Sample:
1; *meso*-Erythritol〔*meso*-Erythrite〕 (10µg)
2; Xylitol (10µg)
3; Palatinose (10µg)
4; Maltitol (10µg)



NACALAI TESQUE, INC

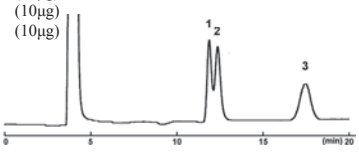
AP-0323

● Infusion Solution Components

COSMOSIL Application Data

Column: Sugar-D
Column size: 4.6mmI.D.-250mm
Mobile phase: Acetonitrile / H_2O = 85/15
Flow rate: 1.0 ml/min
Temperature: 50°C
Detection: RI

Sample: 1; Xylitol (10µg)
2; *D*-(-)-Fructose (10µg)
3; *D*-(+)-Glucose (10µg)



NACALAI TESQUE, INC

AP-0328

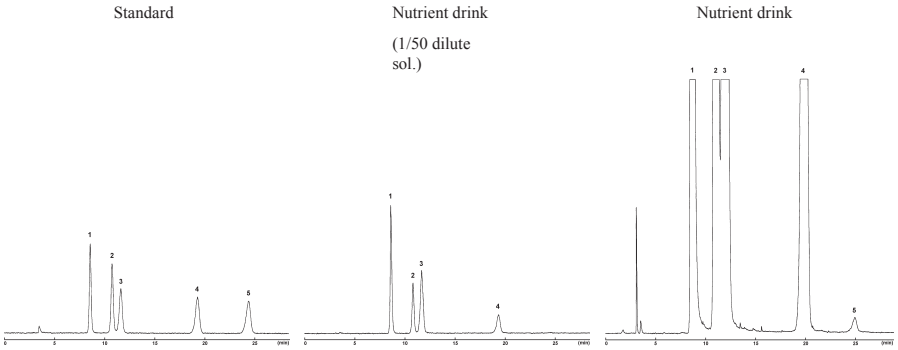
(8) Carbohydrates

● Energy Drink

COSMOSIL Application Data

Column: Sugar-D
Column size: 4.6mmI.D.-250mm
Mobile phase: Acetonitrile/ H_2O = 85/15
Flow rate: 1.0 ml/min
Temperature: 40°C
Detection: ELSD

		Standard
Sample:	1; *D*-(-)-Fructose	(1.0mg/ml)
	2; *D*-(+)-Glucose	(1.0mg/ml)
	3; *D*-Glucitol(Sorbitol)	(1.0mg/ml)
	4; Sucrose	(1.0mg/ml)
	5; *myo*-Inositol	(1.0mg/ml)
Injection Vol.	5.0µl	



NACALAI TESQUE, INC

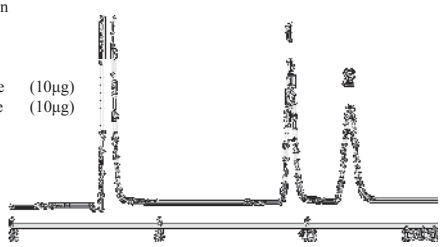
AP-1049

● Chocolate Components

COSMOSIL Application Data

Column: Sugar-D
Column size: 4.6mmI.D.-250mm
Mobile phase: Acetonitrile / H_2O = 75/25
Flow rate: 1.0 ml/min
Temperature: 30°C
Detection: RI

Sample:	1; Sucrose	(10µg)
	2; Lactose	(10µg)



NACALAI TESQUE, INC

AP-0324

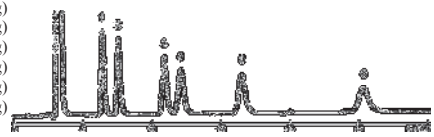
● Gum Components

COSMOSIL Application Data

Column: Sugar-D
Column size: 4.6mmI.D.-250mm
Mobile phase: Acetonitrile / H_2O = 75/25
Flow rate: 1.0 ml/min
Temperature: 30°C
Detection: RI

Sample:

1; Xylitol	(10µg)
2; *D*-(-)-Mannitol	(10µg)
3; Maltitol	(10µg)
4; Palatinit	(10µg)
5; Maltotriitol	(10µg)
6; Maltotertaitol	(10µg)



NACALAI TESQUE, INC

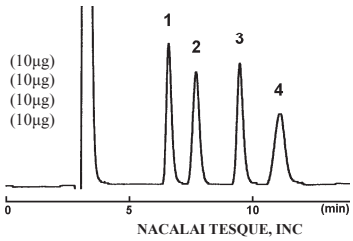
AP-0325

● Cold Beverage Components

COSMOSIL Application Data

Column: Sugar-D
Column size: 4.6mmI.D.-250mm
Mobile phase: Acetonitrile / H_2O = 75/25
Flow rate: 1.0 ml/min
Temperature: 30°C
Detection: RI

Sample:	1; *D*-(-)-Fructose	(10µg)
	2; *D*-(+)-Glucose	(10µg)
	3; Sucrose	(10µg)
	4; Maltose	(10µg)



NACALAI TESQUE, INC

AP-0326

● Sports Drink Components

COSMOSIL Application Data

Column: Sugar-D
Column size: (4.6mmI.D.-250mm) ×2
Mobile phase: Acetonitrile / H_2O = 75/25
Flow rate: 1.0 ml/min
Temperature: 50°C
Detection: RI

Sample:

1; meso-Erythritol 〔meso - Erythrite〕	(10µg)
2; *D*-(-)-Fructose	(10µg)
3; *D*-Glucitol	(10µg)
4; *D*-(+)-Glucose	(10µg)
5; Sucrose	(10µg)
6; Maltose	(10µg)
7; Maltotriitol	(10µg)
8; Maltotriose	(10µg)

NACALAI TESQUE, INC

AP-0327

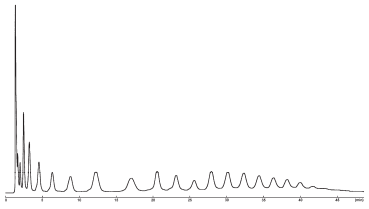
(8) Carbohydrates

● PA–Glucose Oligomer

COSMOSIL Application Data

Column: $5C_{18}$-PAQ
Column size: 4.6mmI.D.-150mm
Mobile phase: A: 20mmol/l Acetate Buffer(pH3.3)
B: 20mmol/l Acetate Buffer(pH3.3)+0.5%Butanol
B(0→100%) 45min Linear gradient
Flow rate: 1.0 ml/min
Temperature: 30°C
Detection: FLS at Ex.320nm
Em. 400nm

Sample:
PA-Glucose Oligomer(DP=3-22)



NACALAI TESQUE, INC

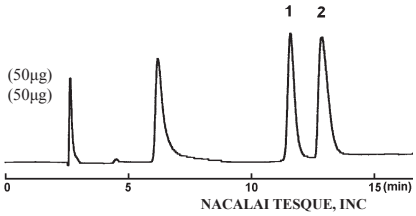
AP-0379

● Phosphorylated Sugars

COSMOSIL Application Data

Column: HILIC
Column size: 4.6mmI.D.-250mm
Mobile phase: Acetonitrile/ 20mmol/l Phosphate Buffer(pH7.0) = 60/40
Flow rate: 1.0 ml/min
Temperature: 30°C
Detection: RI

Sample:
1; *D*-Fructose-*6*-phosphate (50µg)
2; *D*-Glucose-*6*-phosphate (50µg)



NACALAI TESQUE, INC

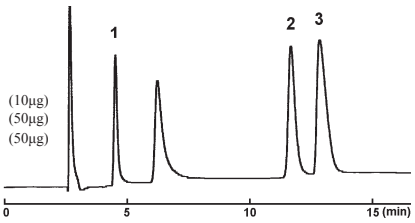
AP-0317

● Phosphorylated Sugars

COSMOSIL Application Data

Column: HILIC
Column size: 4.6mmI.D.-250mm
Mobile phase: Acetonitrile/ 20mmol/l Phosphate Buffer(pH7.0) = 60/40
Flow rate: 1.0 ml/min
Temperature: 30°C
Detection: RI

Sample:
1; *D*-(+)-Glucose (10µg)
2; α-*D*-Glucose-*1*-phosphate (50µg)
3; *D*-Glucose-*6*-phosphate (50µg)



NACALAI TESQUE, INC

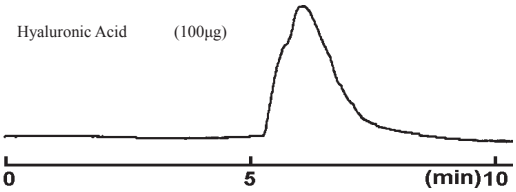
AP-0314

● Hyaluronic Acid

COSMOSIL Application Data

Column: CNT-1000
Column size: 7.5mmI.D.-300mm
Mobile phase: 20mmol/l Phosphate Buffer(pH7),100mmol/l Na_2SO_4
Flow rate: 1.0 ml/min
Temperature: 30°C
Detection: RI

Sample: Hyaluronic Acid (100µg)



NACALAI TESQUE, INC

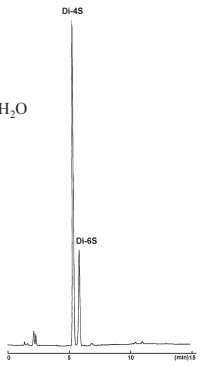
AP-1081

● Enzyme digests of Chondroitin Sulfate A

COSMOSIL Application Data

Column: Cholester
Column size: 4.6mmI.D.-150mm
Mobile phase: A; 1mmol/l Tetrabutylammonium Bisulfate/H_2O
B; 1mmol/l Tetrabutylammonium Bisulfate -Acetonitrile/H_2O=67/33
B conc. 20%→65%(7min)→65%(12min)
Flow rate: 1.0 ml/min
Temperature: 40°C
Detection: UV240nm

Sample: Chondroitin Sulfate A
Chodrotinase AC-II digested



NACALAI TESQUE, INC

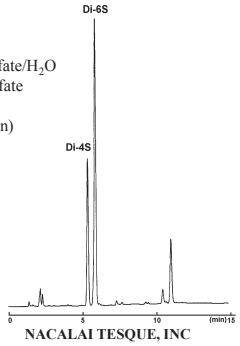
AP-1082

● Enzyme digests of Chondroitin Sulfate C

COSMOSIL Application Data

Column: Cholester
Column size: 4.6mmI.D.-150mm
Mobile phase: A; 1mmol/l Tetrabutylammonium Bisulfate/H_2O
B; 1mmol/l Tetrabutylammonium Bisulfate -Acetonitrile/H_2O=67/33
B conc. 20%→65%(7min)→65%(12min)
Flow rate: 1.0 ml/min
Temperature: 40°C
Detection: UV240nm

Sample: Chondroitin Sulfate C
Chodrotinase AC-II digested



NACALAI TESQUE, INC

AP-1083

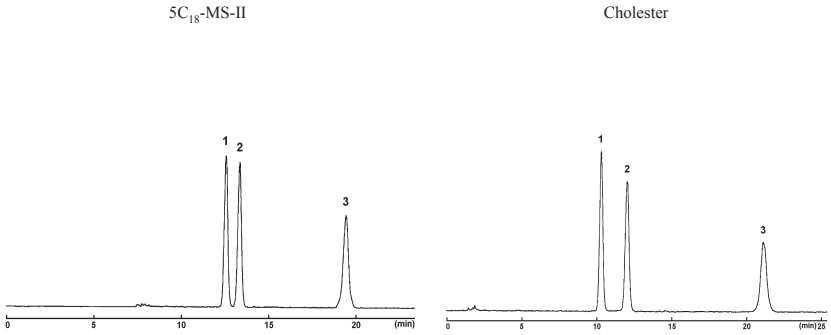
(9) Lipids

● Fatty Acids

COSMOSIL Application Data

Column:
Column size: 4.6mmI.D.-150mm
Mobile phase: 0.05%TFA-90%Methanol
Flow rate: 1.0 ml/min
Temperature: 30°C
Detection: ELSD

Sample: 1; Oleic Acid (3.0µg)
2; Elaidic Acid (3.0µg)
3; Stearic Acid
〔Octadecanoic Acid〕(3.0µg)



NACALAI TESQUE, INC

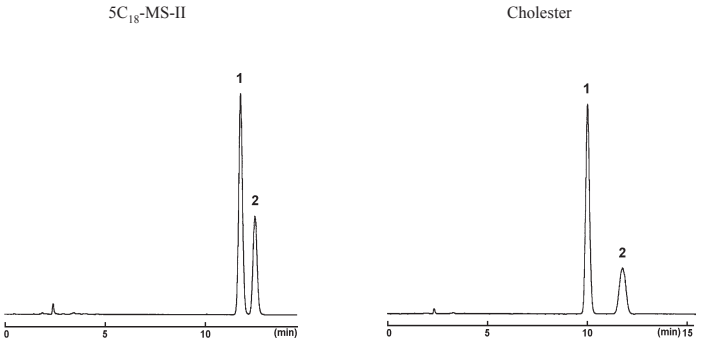
AP-1038

● Fatty Acids

COSMOSIL Application Data

Column:
Column size: 4.6mmI.D.-150mm
Mobile phase: 0.05%TFA-90%Methanol
Flow rate: 1.0 ml/min
Temperature: 30°C
Detection: ELSD

Sample: 1; *cis*-Vaccenic Acid (3.0µg)
2; *trans*-Vaccenic Acid (3.0µg)



NACALAI TESQUE, INC

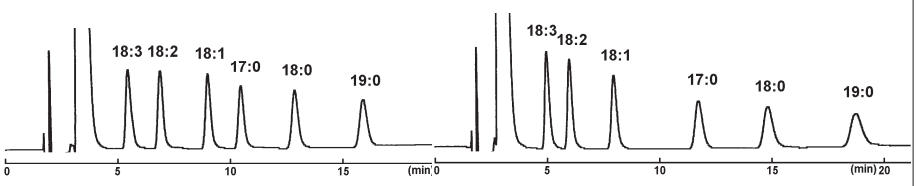
AP-1039

● Methylated Fatty Acids

COSMOSIL Application Data

Column:
Column size: 4.6mmI.D.-150mm
Mobile phase: Methanol/ H_2O = 95/5
Flow rate: 1.0 ml/min
Temperature: 30°C
Detection: RI

Sample: Methyl Linolenate (18:3)
Methyl Linoleate (18:2)
Methyl Oleate (18:1)
Methyl Margarate (17:0)
Methyl Stearate (18:0)
Methyl Nonadecanoate (19:0)
(each 10µg)



NACALAI TESQUE, INC

AP-1040

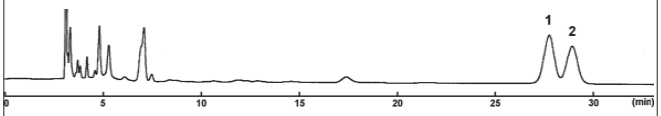
(9) Lipids

● Fatty Acid Derivatives

COSMOSIL Application Data

Column: Cholester
Column size: 4.6mmI.D.-250mm
Mobile phase: Methanol/ H_2O = 90/10
Flow rate: 1.0 ml/min
Temperature: 30°C
Detection: UV254nm

Sample: 1; Linolenic Acid *p*-Bromophenacyl Ester
2; γ-Linolenic Acid *p*-Bromophenacyl Ester



NACALAI TESQUE, INC

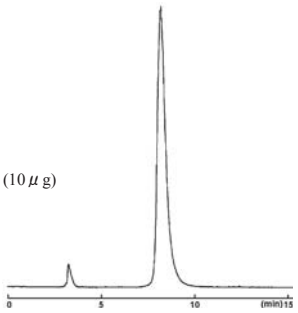
AP-0211

● Phosphatides

COSMOSIL Application Data

Column: 5SL-II
Column size: 4.6mmI.D.-250mm
Mobile phase: 0.05%TFA-Methanol
Flow rate: 1.0 ml/min
Temperature: 30°C
Detection: ELSD

Sample: *L*-α-Phosphatidyl Choline (10μg)



NACALAI TESQUE, INC

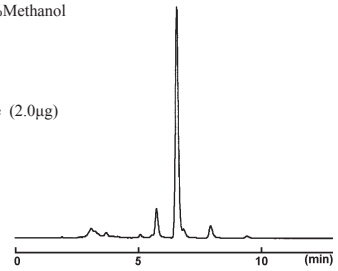
AP-0250

● Phosphatides

COSMOSIL Application Data

Column: $5C_{18}$-MS-II
Column size: 4.6mmI.D.-250mm
Mobile phase: 0.05%TFA-10%THF/90%Methanol
Flow rate: 1.0 ml/min
Temperature: 30°C
Detection: ELSD

Sample: *L*-α-Phosphatidyl Choline (2.0μg)



NACALAI TESQUE, INC

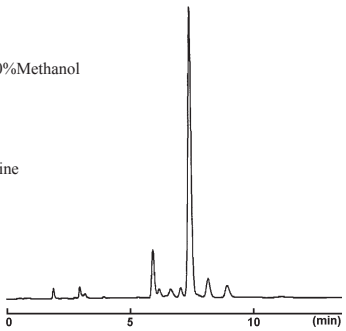
AP-0251

● Phosphatides

COSMOSIL Application Data

Column: $5C_{18}$-MS-II
Column size: 4.6mmI.D.-250mm
Mobile phase: 0.05%TFA-10%THF/90%Methanol
Flow rate: 1.0 ml/min
Temperature: 30°C
Detection: ELSD

Sample: *L*-α-Phosphatidyl-L-serine



NACALAI TESQUE, INC

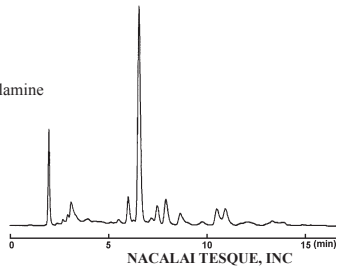
AP-0252

● Phosphatides

COSMOSIL Application Data

Column: $5C_{18}$-MS-II
Column size: 4.6mmI.D.-250mm
Mobile phase: 0.05%TFA-10%THF/90%Methanol
Flow rate: 1.0 ml/min
Temperature: 30°C
Detection: ELSD

Sample: *L*-α-Phosphatidylethanolamine
(8.0μg)



NACALAI TESQUE, INC

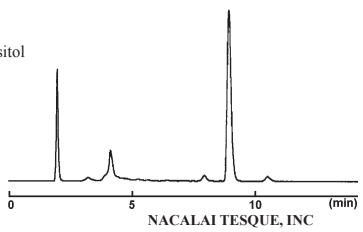
AP-0253

● Phosphatides

COSMOSIL Application Data

Column: $5C_{18}$-MS-II
Column size: 4.6mmI.D.-250mm
Mobile phase: 0.05%TFA-10%THF/90%Methanol
Flow rate: 1.0 ml/min
Temperature: 30°C
Detection: ELSD

Sample: *L*-α-Phosphatidylinositol
Sodium Salt



NACALAI TESQUE, INC

AP-0254

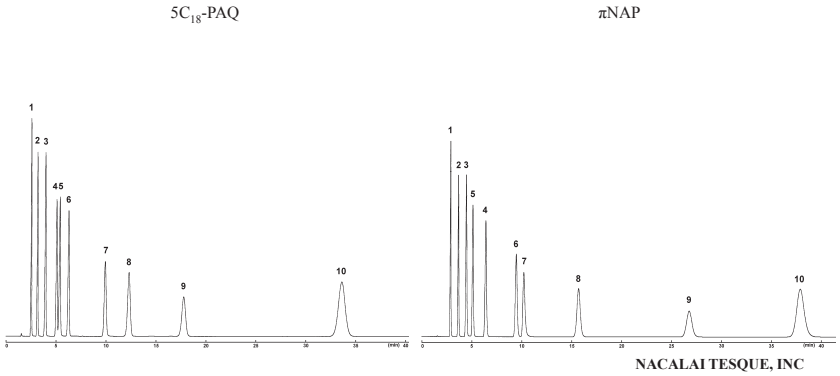
(10) Nucleic Acid Related Substances

● Nucleobases and Nucleosides

COSMOSIL Application Data

Column:
Column size: 4.6mmI.D.-150mm
Mobile phase: 20mmol/l Phosphate Buffer(pH7)
Flow rate: 1.0 ml/min
Temperature: 30°C
Detection: UV260nm

Sample:		
	1; Cytosine	(0.16μg)
	2; Uracil	(0.06μg)
	3; Cytidine	(0.32μg)
	4; Uridine	(0.24μg)
	5; Guanine	(0.16μg)
	6; Thymine	(0.16μg)
	7; Adenine	(0.08μg)
	8; Guanosine	(0.24μg)
	9; Thymidine	(0.24μg)
	10; Adenosine	(0.16μg)



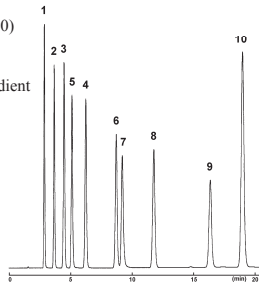
AP-1051

● Nucleobases and Nucleosides

COSMOSIL Application Data

Column: πNAP
Column size: 4.6mmI.D.-150mm
Mobile phase: A; 20mmol/l Phosphate Buffer(pH7.0)
B; Methanol/ 20mmol/l Phosphate Buffer(pH7.0) = 10/90
B conc. 0→100% 15min Linear gradient
Flow rate: 1.0 ml/min
Temperature: 30°C
Detection: UV260nm

Sample:		
	1; Cytosine	(0.16μg)
	2; Uracil	(0.06μg)
	3; Cytidine	(0.32μg)
	4; Uridine	(0.24μg)
	5; Guanine	(0.16μg)
	6; Thymine	(0.16μg)
	7; Adenine	(0.08μg)
	8; Guanosine	(0.24μg)
	9; Thymidine	(0.24μg)
	10; Adenosine	(0.16μg)



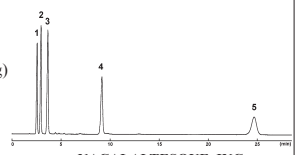
AP-1052

● Nucleobases, Nucleosides and Nucleotides

COSMOSIL Application Data

Column: 5C18-PAQ
Column size: 4.6mmI.D.-150mm
Mobile phase: 20mmol/l Phosphate Buffer(pH7)
Flow rate: 1.0 ml/min
Temperature: 30°C
Detection: UV260nm

Sample:		
	1; Adenosine-5′-triphosphate	(0.2μg)
	2; Adenosine-5′-diphosphate	(0.2μg)
	3; Adenosine-5′-monophosphate	(0.2μg)
	4; Adenine	(0.05μg)
	5; Adenosine	(0.1μg)



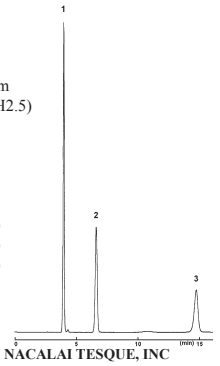
AP-0376

● Nucleotides

COSMOSIL Application Data

Column: 5C18-PAQ
Column size: 4.6mmI.D.-250mm
Mobile phase: Methanol/ 5mmol/l Tetra-*n*-butylammonium Phosphate, 20mmol/l Phosphate Buffer (pH2.5) = 20/80
Flow rate: 1.0 ml/min
Temperature: 30°C
Detection: UV260nm

Sample:		
	1; Adenosine-5′-monophosphate	(0.33μg)
	2; Adenosine-5′-diphosphate	(0.33μg)
	3; Adenosine-5′-triphosphate	(0.33μg)



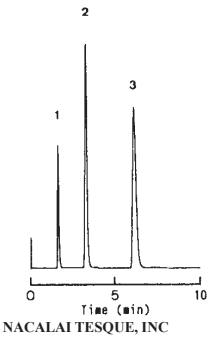
AP-0377

● Nucleotides

COSMOSIL Application Data

Column: 5C18-AR-II
Column size: 4.6mmI.D.-150mm
Mobile phase: 20mmol/l Phosphate Buffer(pH7)
Flow rate: 1.0 ml/min
Temperature: 30°C
Detection: UV254nm, 0.5AUFS

Sample:
1; 2′-Deoxycytidine 5′-Monophosphate [dCMP] (1.0μg)
2; 2′-Deoxyguanosine 5′-Monophosphate [dGMP](1.2μg)
3; 2′-Deoxyadenosine-5′-Monophosphate [dAMP] (1.0μg)



AP-0115

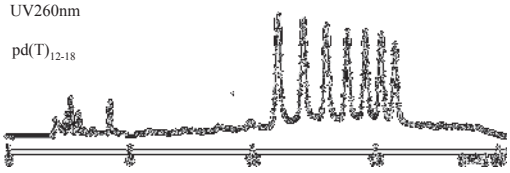
(10) Nucleic Acid Related Substances

● Oligonucleotide

COSMOSIL Application Data

Column: Protein-R
Column size: 4.6mmI.D.-150mm
Mobile phase: A; 100mmol/l Ammonium Acetate
B; Acetonitrile
B conc. 9→11% 15min Linear gradient
Flow rate: 1.0 ml/min
Temperature: 30°C
Detection: UV260nm

Sample: $pd(T)_{12-18}$



NACALAI TESQUE, INC

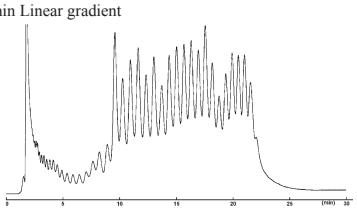
AP-1064

● Oligonucleotide

COSMOSIL Application Data

Column: COSMOGEL IEX Type Q-N
Column size: 4.6mmI.D.-100mm
Mobile phase: A: 20mmol/l Tris Buffer(pH8.3)
B: 20mmol/l Tris Buffer(pH8.3)
+ 1.0mol/l NaCl
B conc. 50→60% 30min Linear gradient
Flow rate: 0.5 ml/min
Temperature: 30°C
Detection: UV260nm

Sample: DNA〔pd(A)40-60〕



NACALAI TESQUE, INC

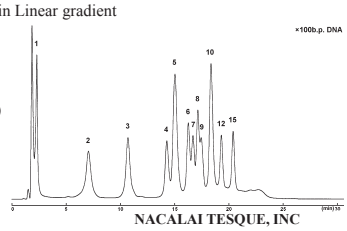
AP-1170

● DNA Ladder

COSMOSIL Application Data

Column: COSMOGEL IEX Type Q-N
Column size: 4.6mmI.D.-100mm
Mobile phase: A: 20mmol/l Tris Buffer(pH8.3)
B: 20mmol/l Tris Buffer(pH8.3)
+ 1.0mol/l NaCl
B conc. 75→85% 30min Linear gradient
Flow rate: 0.5 ml/min
Temperature: 30°C
Detection: UV260nm

Sample: DNA (100b.p. Ladder)



NACALAI TESQUE, INC

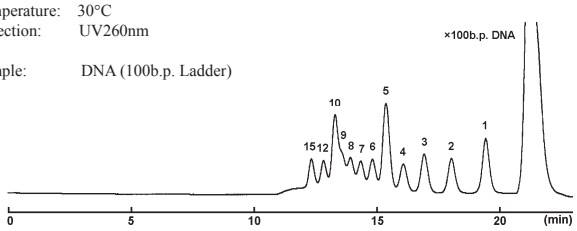
AP-1169

● DNA Ladder

COSMOSIL Application Data

Column: CNT-1000 + CNT-2000
Column size: 7.5mmI.D.-300mm × 2
Mobile phase: 20mmol/l Phosphate Buffer(pH7),100mmol/l Na_2SO_4
Flow rate: 1.0 ml/min
Temperature: 30°C
Detection: UV260nm

Sample: DNA (100b.p. Ladder)



NACALAI TESQUE, INC

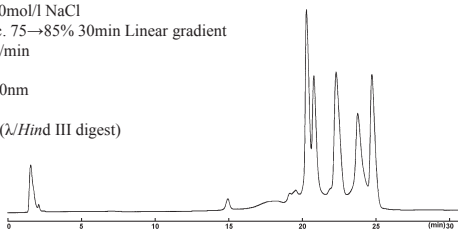
AP-1078

● λDNA/Hind III digest

COSMOSIL Application Data

Column: COSMOGEL IEX Type Q-N
Column size: 4.6mmI.D.-100mm
Mobile phase: A: 20mmol/l Tris buffer(pH8.3)
B: 20mmol/l Tris buffer(pH8.3)
+ 1.0mol/l NaCl
B conc. 75→85% 30min Linear gradient
Flow rate: 0.5 ml/min
Temperature: 30°C
Detection: UV260nm

Sample: DNA (λ/*Hin*d III digest)



NACALAI TESQUE, INC

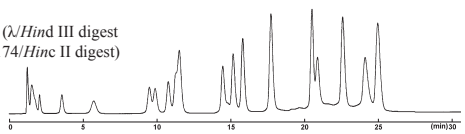
AP-1171

● λDNA/Hind III digest–ØX174/Hinc II digest

COSMOSIL Application Data

Column: COSMOGEL IEX Type Q-N
Column size: 4.6mmI.D.-100mm
Mobile phase: A: 20mmol/l Tris buffer(pH8.3)
B: 20mmol/l Tris buffer(pH8.3)
+ 1.0mol/l NaCl
B conc. 75→85% 30min Linear gradient
Flow rate: 0.5 ml/min
Temperature: 30°C
Detection: UV260nm

Sample: DNA (λ/*Hin*d III digest
- φX174/*Hin*c II digest)



NACALAI TESQUE, INC

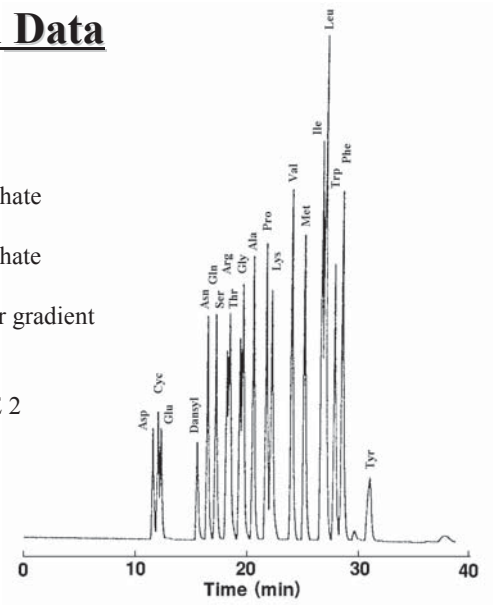
AP-1172

● Dansyl Amino Acids

COSMOSIL Application Data

Column: $5C_{18}$-MS-II
Column size: 4.6mmI.D.-150mm
Mobile phase: A; Acetonitrile/ 20mmol Phosphate buffer(pH7.0) =10/90
B; Acetonitrile/ 20mmol Phosphate buffer(pH7.0) =40/60
B conc. 0→100% 30min Linear gradient
Flow rate: 1.0 ml/min
Temperature: 30°C
Detection: Ex.365nm Em.530nm RANGE 2

Sample: Dansyl Amino Acids (1.0 μg each)



NACALAI TESQUE, INC

AP-0003

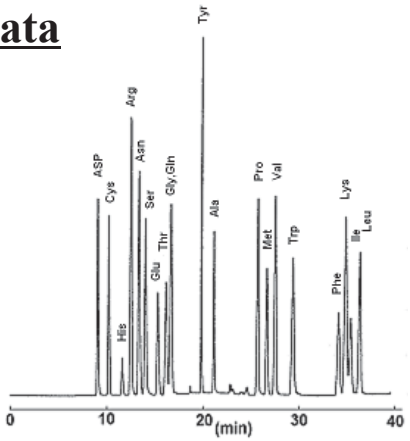
● PTH–Amino Acids

COSMOSIL Application Data

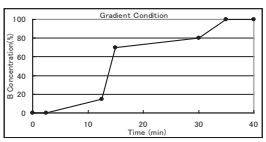
Column: $5C_{18}$-AR-II
Column size: 4.6mmI.D.-150mm
Mobile phase: A; Acetonitrile/ 20mmol/l Acetate buffer(pH4.8) =10/90
B; Acetonitrile/ 20mmol/l Acetate buffer(pH4.8) =50/50
Step wise gradient
Flow rate: 1.0 ml/min
Temperature: 60°C
Detection: UV270nm, 0.16AUFS
Sample:

PTH-Asp	(0.4mg/ml)
PTH-His	(0.3mg/ml)
PTH-Arg	(0.5mg/ml)
PTH-Ser	(0.3mg/ml)
PTH-Trp	(0.25mg/ml)
PTH-Lys	(0.35mg/ml)
Others	(0.2mg/ml)

Injection 2.0 μl



Gradient Condition	
Time (min)	B Con.(%)
0	0
2.5	0
12.5	15
15	70
30	80
35	100
40	100



NACALAI TESQUE, INC

AP-0004

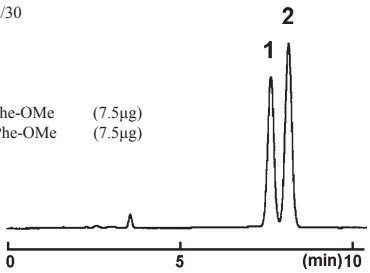
(11) Amino Acids, Peptides and Proteins

● Amino–Acid Derivatives (Diastereomer)

COSMOSIL Application Data

Column: $5C_{18}$-MS-II
Column size: 4.6mmI.D.-150mm
Mobile phase: Methanol/ H_2O = 70/30
Flow rate: 1.0 ml/min
Temperature: 30°C
Detection: UV254nm

Sample: 1; Boc-*(L)*Phe-*(L)*Phe-OMe (7.5µg)
2; Boc-*(D)*Phe-*(L)*Phe-OMe (7.5µg)



NACALAI TESQUE, INC

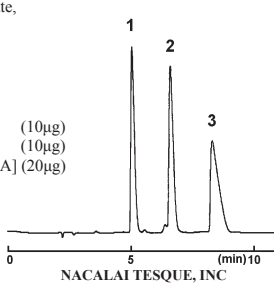
AP-0241

● The umami of Vesitables

COSMOSIL Application Data

Column: $5C_{18}$-PAQ
Column size: 4.6mmI.D.-250mm
Mobile phase: 5mmol/l Sodium *1*-Hexanesulfonate,
20mmol/l Phosphoric Acid
Flow rate: 1.0 ml/min
Temperature: 30°C
Detection: UV210nm

Sample: 1; *L*-Aspartic Acid (10µg)
2; *L*-Glutamic Acid (10µg)
3; *4*-Amino-*n*-butyric Acid [GABA] (20µg)



NACALAI TESQUE, INC

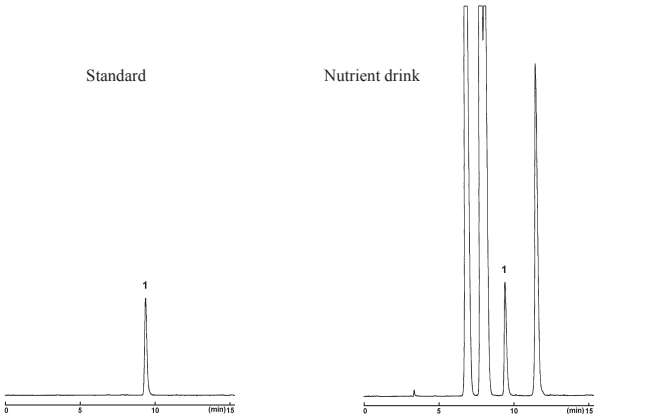
AP-1015

● Energy Drink

COSMOSIL Application Data

Column: HILIC
Column size: 4.6mmI.D.-250mm
Mobile phase: Acetonitrile/ 10mmol/l
Ammonium Acetate = 80/20
Flow rate: 1.0 ml/min
Temperature: 30°C
Detection: ELSD

Standard
Sample: 1; Taurine (10mg/ml)
Injection Vol. 0.5µl



NACALAI TESQUE, INC

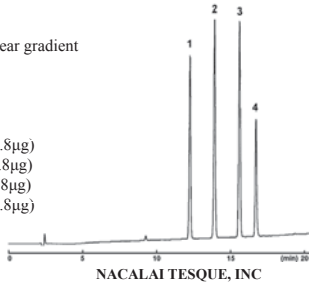
AP-1050

● Peptides

COSMOSIL Application Data

Column: Protein-R
Column size: 4.6mmI.D.-150mm
Mobile phase: A; 0.05%TFA-H_2O
B; 0.05%TFA-Acetonitrile
B conc. 10→40% 20min Linear gradient
Flow rate: 1.0 ml/min
Temperature: 30°C
Detection: UV220nm

Sample: 1; Oxytocin (0.8µg)
2; Angiotensin II(Human) (0.8µg)
3; Angiotensin I(Human) (0.8µg)
4; Substance P (0.8µg)



NACALAI TESQUE, INC

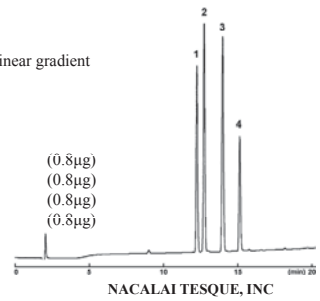
AP-0350

● Peptides

COSMOSIL Application Data

Column: Cholester
Column size: 4.6mmI.D.-150mm
Mobile phase: A; 0.05%TFA-H_2O
B; 0.05%TFA-Acetonitrile
B conc. 10→40% 20min Linear gradient
Flow rate: 1.0 ml/min
Temperature: 30°C
Detection: UV220nm

Sample: 1; Oxytocin (0.8µg)
2; Angiotensin II(Human) (0.8µg)
3; Angiotensin I(Human) (0.8µg)
4; Substance P (0.8µg)



NACALAI TESQUE, INC

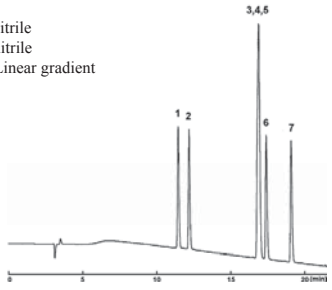
AP-0238

(11) Amino Acids, Peptides and Proteins

● Peptides

COSMOSIL Application Data

Column: $5C_{18}$-AR-II
Column size: 4.6mmI.D.-250mm
Mobile phase: A; 0.05%TFA-10%Acetonitrile
B; 0.05%TFA-30%Acetonitrile
B conc. 0→100% 20min Linear gradient
Flow rate: 1.0 ml/min
Temperature: 30°C
Detection: UV220nm
Sample:
1; Angiotensin II, [Sar^1,Thr^8] (0.49µg)
2; Angiotensin II, [Sar^1,Ala^8] (0.49µg)
3; Angiotensin II, Des-Asp^1-[Ile^8] (0.49µg)
4; Angiotensin II, [Sar^1,Ile^8] (0.49µg)
5; Angiotensin II, [Asn^1,Val^5] (0.49µg)
6; Angiotensin II, [Val^5] (0.49µg)
7; Angiotensin II (Human) (0.49µg)



NACALAI TESQUE, INC

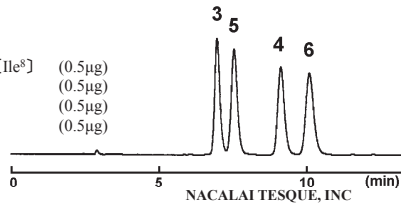
AP-1005

● Peptides

COSMOSIL Application Data

Column: HILIC
Column size: 4.6mmI.D.-250mm
Mobile phase: Acetonitrile/ 10mmol/l Phosphate Buffer(pH7.0) = 70/30
Flow rate: 1.0 ml/min
Temperature: 30°C
Detection: UV220nm

Sample:
3; Angiotensin II, Des-Asp1-[Ile^8] (0.5µg)
4; Angiotensin II, [Sar^1,Ile^8] (0.5µg)
5; Angiotensin II, [Asn^1,Val^5] (0.5µg)
6; Angiotensin II, [Val^5] (0.5µg)



NACALAI TESQUE, INC

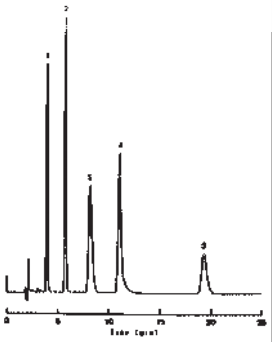
AP-0316

● Peptides

COSMOSIL Application Data

Column: $5C_{18}$-AR-300
Column size: 4.6mmI.D.-150mm
Mobile phase: 0.05%TFA-22%Acetonitrile
Flow rate: 1.0 ml/min
Temperature: 30°C
Detection: UV220nm, 0.16AUFS

Sample:
1; Bradykinin (1.0µg)
2; Angiotensin II (1.0µg)
3; Neurotensin (1.0µg)
4; Bombesin (1.0µg)
5; Substance P (1.0µg)



NACALAI TESQUE, INC

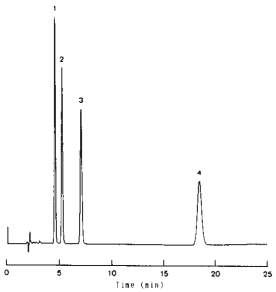
AP-0009

● Peptides

COSMOSIL Application Data

Column: $5C_{18}$-AR-300
Column size: 4.6mmI.D.-150mm
Mobile phase: 0.05%TFA-22%Acetonitrile
Flow rate: 1.0 ml/min
Temperature: 30°C
Detection: UV220nm, 0.16AUFS

Sample:
1; Methionine-Enkephalin (0.5µg)
2; [Ala^2]-Methionine-Enkephalin (0.5µg)
3; Leucine-Enkephalin (0.5µg)
4; [Ala^2]-Leucine-Enkephalin (0.5µg)



NACALAI TESQUE, INC

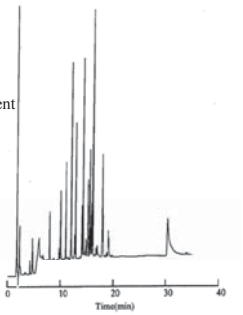
AP-0005

● Peptide Mappings

COSMOSIL Application Data

Column: $5C_{18}$-AR-II
Column size: 4.6mmI.D.-150mm
Mobile phase: A; 5mmol/l TFA-H_2O
B; 5mmol/l TFA-60%Acetonitrile
B conc. 0→100% 30min Linear gradient
Flow rate: 1.0 ml/min
Temperature: 30°C
Detection: UV210nm, 0.32AUFS

Sample: Cytochrome C
Lysyl Endopeptidase digested
(2.5µl)



NACALAI TESQUE, INC

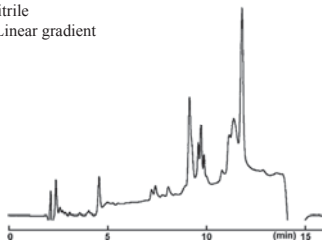
AP-0002

● Semi-purified Myosin

COSMOSIL Application Data

Column: Protein-R
Column size: 4.6mmI.D.-150mm
Mobile phase: A:0.05%TFA-20%Acetonitrile
B:0.05%TFA-60%Acetonitrile
B conc. 0→100% 10min Linear gradient
Flow rate: 1.0 ml/min
Temperature: 30°C
Detection: UV220nm, 0.256AUFS

Sample: Myosin (20µg)



NACALAI TESQUE, INC

AP-0346

(11) Amino Acids, Peptides and Proteins

● Milk Protein

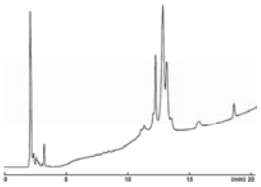
COSMOSIL Application Data

Column: Protein-R
Column size: 4.6mmI.D.-150mm
Mobile phase: A; 0.05%TFA-H_2O
B; 0.05%TFA-Acetonitrile
B conc. 20→80% 20min Linear gradient
Flow rate: 1.0 ml/min
Temperature: 30°C
Detection: UV220nm

Sample: Milk

Sample Preparation:
・Ultracentrifuged at 90,000 g for 1 hr.
・Clear supernatant solution was injected.

Injection vol.: 1.0µl



NACALAI TESQUE, INC

AP-1079

● Soymilk Protein

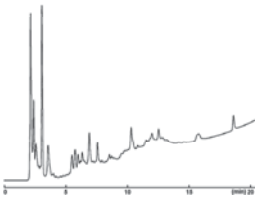
COSMOSIL Application Data

Column: Protein-R
Column size: 4.6mmI.D.-150mm
Mobile phase: A; 0.05%TFA-H_2O
B; 0.05%TFA-Acetonitrile
B conc. 20→80% 20min Linear gradient
Flow rate: 1.0 ml/min
Temperature: 30°C
Detection: UV220nm

Sample: Soybean milk

Sample Preparation:
・Ultracentrifuged at 90,000 g for 1 hr.
・Clear supernatant solution was injected.

Injection vol.: 1.0µl



NACALAI TESQUE, INC

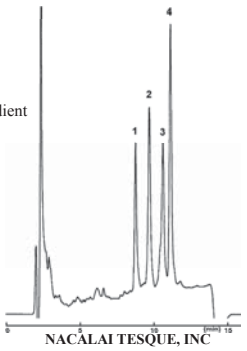
AP-1080

● Bacteria-derived Proteins

COSMOSIL Application Data

Column: Protein-R
Column size: 4.6mmI.D.-150mm
Mobile phase: A:0.05%TFA-20%Acetonitrile
B:0.05%TFA-60%Acetonitrile
B conc. 0→100% 10min Linear gradient
Flow rate: 1.0 ml/min
Temperature: 30°C
Detection: UV220nm, 0.256AUFS

Sample: 1; Choline Oxidase (6.0µg)
2; α-Amylase (3.0µg)
3; Glucose Oxidase (6.0µg)
4; Thermolysin (9.0µg)



NACALAI TESQUE, INC

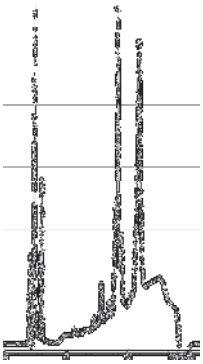
AP-0337

● Bacteria-derived Proteins

COSMOSIL Application Data

Column: Protein-R
Column size: 4.6mmI.D.-150mm
Mobile phase: A:0.05%TFA-20%Acetonitrile
B:0.05%TFA-60%Acetonitrile
B conc. 0→100% 10min Linear gradient
Flow rate: 1.0 ml/min
Temperature: 30°C
Detection: UV220nm, 0.256AUFS

Sample: 1; Actinase E (13.4µg)
2; Alcohol Dehydrogenase (6.6µg)



NACALAI TESQUE, INC

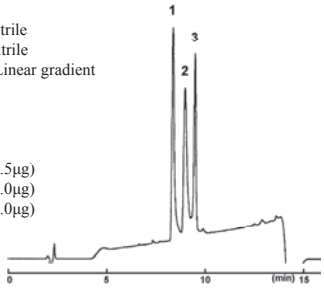
AP-0338

● Human-derived Proteins

COSMOSIL Application Data

Column: Protein-R
Column size: 4.6mmI.D.-150mm
Mobile phase: A:0.05%TFA-20%Acetonitrile
B:0.05%TFA-60%Acetonitrile
B conc. 0→100% 10min Linear gradient
Flow rate: 1.0 ml/min
Temperature: 30°C
Detection: UV220nm, 0.256AUFS

Sample: 1; Transferrin (1.5µg)
2; Albumin, Human (2.0µg)
3; Carbonic Anhydrase (1.0µg)



NACALAI TESQUE, INC

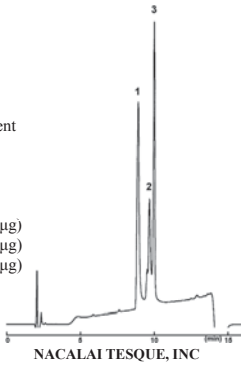
AP-0339

● Bovine-derived Proteins

COSMOSIL Application Data

Column: Protein-R
Column size: 4.6mmI.D.-150mm
Mobile phase: A:0.05%TFA-20%Acetonitrile
B:0.05%TFA-60%Acetonitrile
B conc. 0→100% 10min Linear gradient
Flow rate: 1.0 ml/min
Temperature: 30°C
Detection: UV220nm, 0.256AUFS

Sample: 1; Albumin, Bovine [BSA] (2.0µg)
2; *L*-Glutamic Dehydrogenase (1.5µg)
3; Carbonic Anhydrase (1.5µg)



NACALAI TESQUE, INC

AP-0340

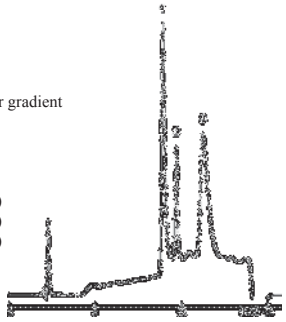
(11) Amino Acids, Peptides and Proteins

● Bovine-derived Proteins

COSMOSIL Application Data

Column: Protein-R
Column size: 4.6mmI.D.-150mm
Mobile phase: A:0.05%TFA-20%Acetonitrile
B:0.05%TFA-60%Acetonitrile
B conc. 0→100% 10min Linear gradient
Flow rate: 1.0 ml/min
Temperature: 30°C
Detection: UV220nm, 0.256AUFS

Sample: 1; Fibrinogen (4.0µg)
2; Catalase (2.0µg)
3; Thyroglobulin (6.0µg)



NACALAI TESQUE, INC

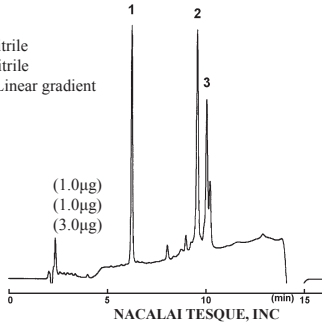
AP-0341

● Bovine Spleen-derived Proteins

COSMOSIL Application Data

Column: Protein-R
Column size: 4.6mmI.D.-150mm
Mobile phase: A:0.05%TFA-20%Acetonitrile
B:0.05%TFA-60%Acetonitrile
B conc. 0→100% 10min Linear gradient
Flow rate: 1.0 ml/min
Temperature: 30°C
Detection: UV220nm, 0.256AUFS

Sample: 1; Ribonuclease A (1.0µg)
2; α-Chymotrypsinogen A (1.0µg)
3; Deoxyribonuclease I (3.0µg)



NACALAI TESQUE, INC

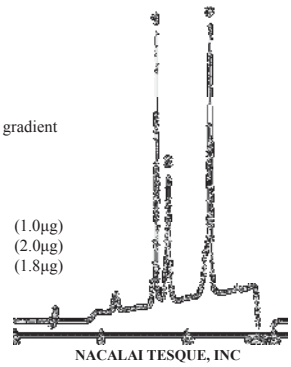
AP-0342

● Bovine Spleen-derived Proteins

COSMOSIL Application Data

Column: Protein-R
Column size: 4.6mmI.D.-150mm
Mobile phase: A:0.05%TFA-20%Acetonitrile
B:0.05%TFA-60%Acetonitrile
B conc. 0→100% 10min Linear gradient
Flow rate: 1.0 ml/min
Temperature: 30°C
Detection: UV220nm, 0.256AUFS

Sample: 1; Lysozyme (1.0µg)
2; Conalbumin (2.0µg)
3; Albumin, Egg〔Ovalbumin〕 (1.8µg)



NACALAI TESQUE, INC

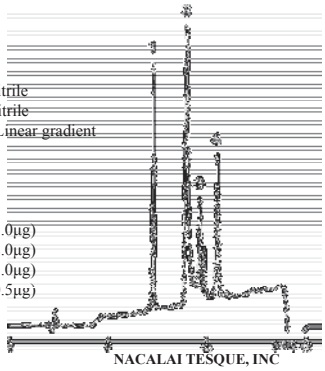
AP-0343

● Other Proteins

COSMOSIL Application Data

Column: Protein-R
Column size: 4.6mmI.D.-150mm
Mobile phase: A:0.05%TFA-20%Acetonitrile
B:0.05%TFA-60%Acetonitrile
B conc. 0→100% 10min Linear gradient
Flow rate: 1.0 ml/min
Temperature: 30°C
Detection: UV220nm, 0.256AUFS

Sample: 1; Cytochrome C (1.0µg)
2; Albumin, Goat (2.0µg)
3; Myoglobin (1.0µg)
4; Concanavalin A (0.5µg)



NACALAI TESQUE, INC

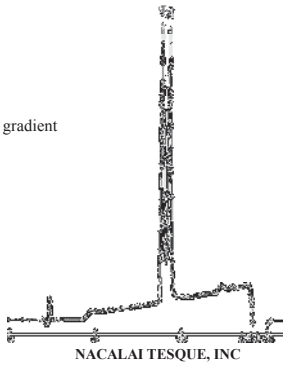
AP-0344

● Other Proteins

COSMOSIL Application Data

Column: Protein-R
Column size: 4.6mmI.D.-150mm
Mobile phase: A:0.05%TFA-20%Acetonitrile
B:0.05%TFA-60%Acetonitrile
B conc. 0→100% 10min Linear gradient
Flow rate: 1.0 ml/min
Temperature: 30°C
Detection: UV220nm, 0.256AUFS

Sample: 1; Albumin, Rabbit (3.2µg)
2; Peroxidase (3.2µg)



NACALAI TESQUE, INC

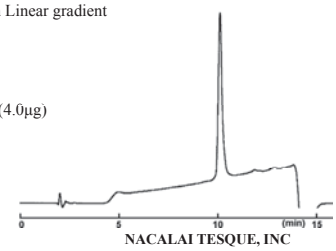
AP-0345

● Pyruvate Kinase

COSMOSIL Application Data

Column: Protein-R
Column size: 4.6mmI.D.-150mm
Mobile phase: A:0.05%TFA-20%Acetonitrile
B:0.05%TFA-60%Acetonitrile
B conc. 0→100% 10min Linear gradient
Flow rate: 1.0 ml/min
Temperature: 30°C
Detection: UV220nm, 0.256AUFS

Sample: Pyruvate Kinase (4.0µg)



NACALAI TESQUE, INC

AP-0348

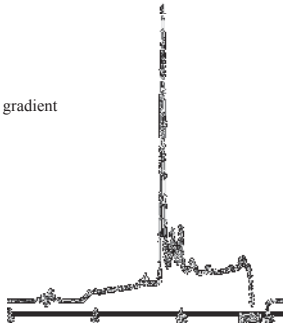
(11) Amino Acids, Peptides and Proteins

● Semi-purified Diaphorase

COSMOSIL Application Data

Column: Protein-R
Column size: 4.6mmI.D.-150mm
Mobile phase: A:0.05%TFA-20%Acetonitrile
B:0.05%TFA-60%Acetonitrile
B conc. 0→100% 10min Linear gradient
Flow rate: 1.0 ml/min
Temperature: 30°C
Detection: UV220nm, 0.256AUFS

Sample: Diaphorase (6.0µg)



NACALAI TESQUE, INC

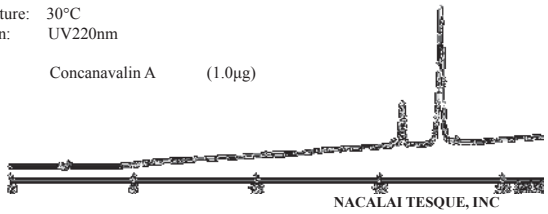
AP-0347

● Glycoproteins

COSMOSIL Application Data

Column: Protein-R
Column size: 4.6mmI.D.-150mm
Mobile phase: A; 0.05%TFA-20%Acetonitrile
B; 0.05%TFA-60%Acetonitrile
B conc. 0→100% 20min Linear gradient
Flow rate: 1.0 ml/min
Temperature: 30°C
Detection: UV220nm

Sample: Concanavalin A (1.0µg)



NACALAI TESQUE, INC

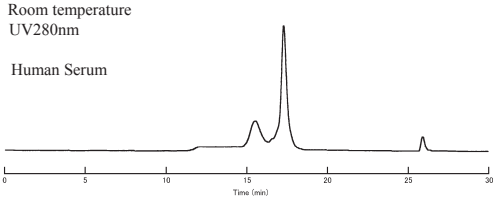
AP-0352

● Human Serum

COSMOSIL Application Data

Column: 5Diol-300-II
Column size: 7.5mmI.D.-600mm
Mobile phase: 20mmol/l Phosphate Buffer(pH 7.0)
＋100mmol/l Na_2SO_4
Flow rate: 1.0 ml/min
Temperature: Room temperature
Detection: UV280nm

Sample: Human Serum



NACALAI TESQUE, INC

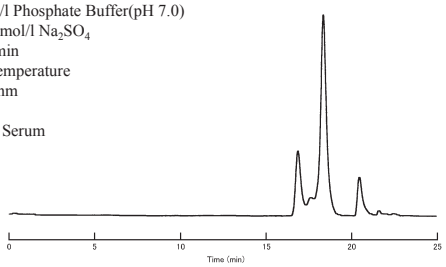
AP-0383

● Bovine Serum

COSMOSIL Application Data

Column: 5Diol-300-II
Column size: 7.5mmI.D.-600mm
Mobile phase: 20mmol/l Phosphate Buffer(pH 7.0)
＋100mmol/l Na_2SO_4
Flow rate: 1.0 ml/min
Temperature: Room temperature
Detection: UV280nm

Sample: Bovine Serum



NACALAI TESQUE, INC

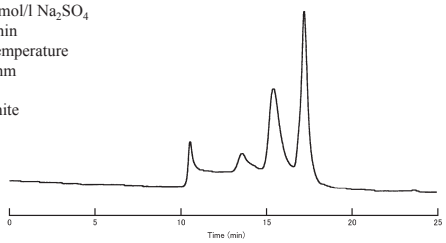
AP-0387

● Egg White

COSMOSIL Application Data

Column: 5Diol-300-II
Column size: 7.5mmI.D.-600mm
Mobile phase: 20mmol/l Phosphate Buffer(pH 7.0)
＋100mmol/l Na_2SO_4
Flow rate: 1.0 ml/min
Temperature: Room temperature
Detection: UV280nm

Sample: Egg White



NACALAI TESQUE, INC

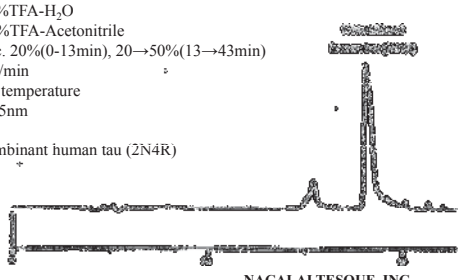
AP-0385

● Tau Protein

COSMOSIL Application Data

Column: Protein-R
Column size: 20mmI.D.-250mm
Mobile phase: A; 0.1%TFA-H_2O
B; 0.1%TFA-Acetonitrile
B conc. 20%(0-13min), 20→50%(13→43min)
Flow rate: 9.0 ml/min
Temperature: Room temperature
Detection: UV215nm

Sample: Recombinant human tau (2N4R)



NACALAI TESQUE, INC

Data courtesy of Akihiko Takashima, Ph.D.
Laboratory for Alzheimer's Disease, RIKEN Brain Science Institute

AP-1150

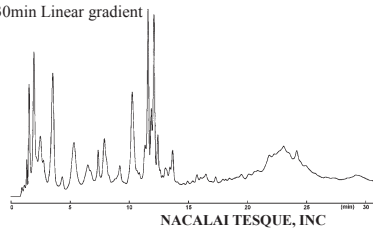
(11) Amino Acids, Peptides and Proteins

● E. coli Proteins

COSMOSIL Application Data

Column: COSMOGEL IEX Type Q
Column size: 4.6mmI.D.-50mm
Mobile phase: A: 20mmol/l Tris Buffer(pH8.3)
B: 20mmol/l Tris Buffer(pH8.3)
+ 0.5mol/l NaCl
B conc. 0→100% 30min Linear gradient
Flow rate: 0.5 ml/min
Temperature: 30°C
Detection: UV220nm

Sample: E.coli Proteins



NACALAI TESQUE, INC

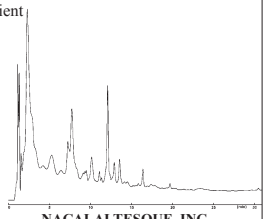
AP-1158

● HeLa Cell Proteins

COSMOSIL Application Data

Column: COSMOGEL IEX Type Q
Column size: 4.6mmI.D.-50mm
Mobile phase: A: 20mmol/l Tris Buffer(pH8.3)
B: 20mmol/l Tris Buffer(pH8.3)
+ 0.5mol/l NaCl
B conc. 0→100% 30min Linear gradient
Flow rate: 0.5 ml/min
Temperature: 30°C
Detection: UV220nm

Sample: HeLa Cell Proteins



NACALAI TESQUE, INC

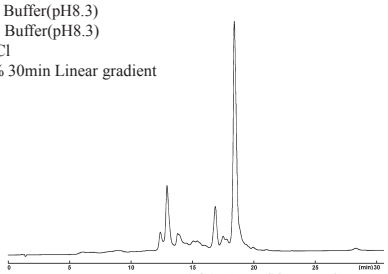
AP-1159

● Egg White

COSMOSIL Application Data

Column: COSMOGEL IEX Type Q
Column size: 4.6mmI.D.-50mm
Mobile phase: A: 20mmol/l Tris Buffer(pH8.3)
B: 20mmol/l Tris Buffer(pH8.3)
+ 0.5mol/l NaCl
B conc. 0→100% 30min Linear gradient
Flow rate: 0.5 ml/min
Temperature: 30°C
Detection: UV220nm

Sample: Egg White



NACALAI TESQUE, INC

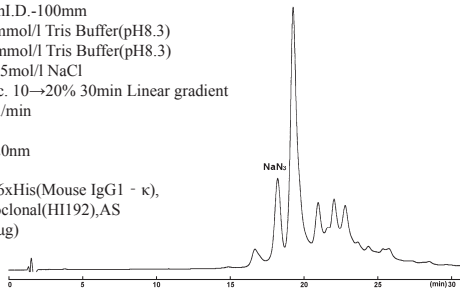
AP-1160

● IgG

COSMOSIL Application Data

Column: COSMOGEL IEX Type Q-N
Column size: 4.6mmI.D.-100mm
Mobile phase: A: 20mmol/l Tris Buffer(pH8.3)
B: 20mmol/l Tris Buffer(pH8.3)
+ 0.5mol/l NaCl
B conc. 10→20% 30min Linear gradient
Flow rate: 0.5 ml/min
Temperature: 30°C
Detection: UV220nm

Sample: Anti-6xHis(Mouse IgG1 - κ), Monoclonal(HI192),AS (10.5µg)



NACALAI TESQUE, INC

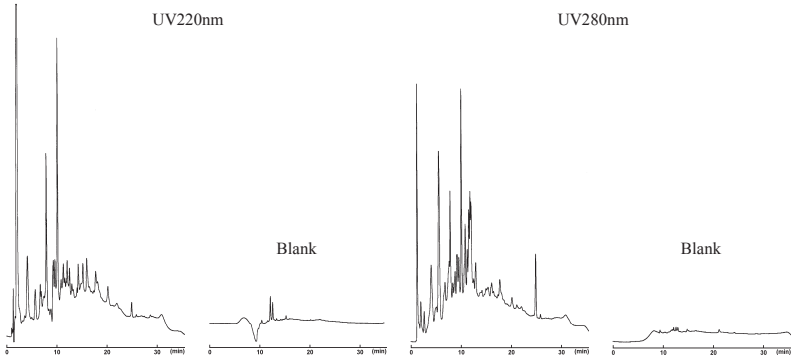
AP-1163

● Urease

COSMOSIL Application Data

Column: COSMOGEL IEX Type M
Column size: 4.6mmI.D.-50mm
Mobile phase: A: 20mmol/l Tris Buffer(pH8.3)
B: 20mmol/l Tris Buffer(pH8.3) + 0.5mol/l NaCl
B conc. 0→100% 30min Linear gradient
Flow rate: 0.5 ml/min
Temperature: 30°C
Detection:

Sample: Urease (100µg)



NACALAI TESQUE, INC

AP-1186

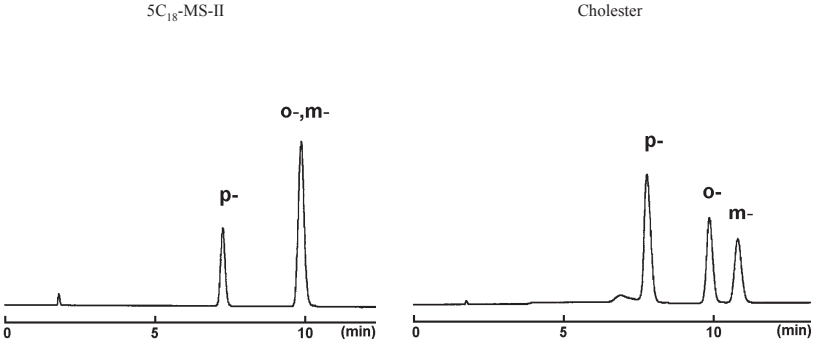
(12) The Others

● Methoxyphenols

COSMOSIL Application Data

Column:
Column size: 4.6mmI.D.-150mm
Mobile phase: Methanol/ H_2O = 30/70
Flow rate: 1.0 ml/min
Temperature: 30°C
Detection: UV254nm

Sample: *o*-Methoxyphenol [Guaiacol] (3.3µg)
m-Methoxyphenol (3.3µg)
p-Methoxyphenol (3.3µg)



NACALAI TESQUE, INC

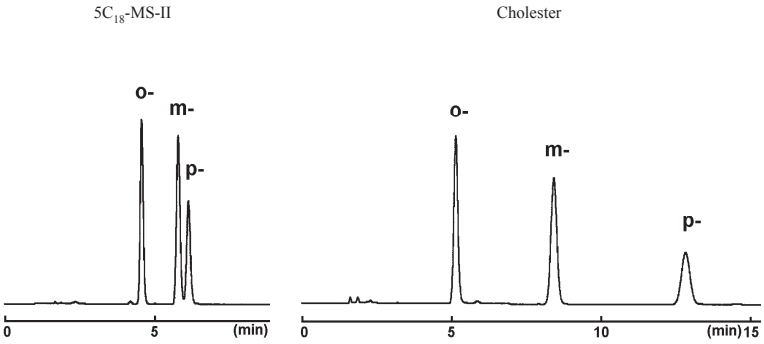
AP-1041

● Terphenyls

COSMOSIL Application Data

Column:
Column size: 4.6mmI.D.-150mm
Mobile phase: Methanol/ H_2O = 90/10
Flow rate: 1.0 ml/min
Temperature: 30°C
Detection: UV254nm

Sample: *o*-Terphenyl (0.15µg)
m-Terphenyl (0.05µg)
p-Terphenyl (0.075µg)



NACALAI TESQUE, INC

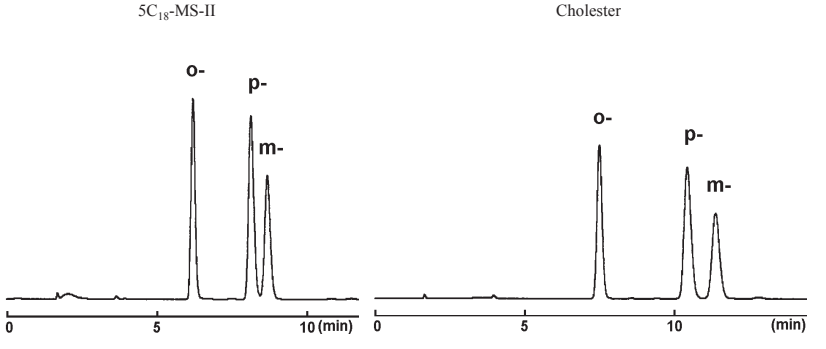
AP-1042

● Chlorophenols

COSMOSIL Application Data

Column:
Column size: 4.6mmI.D.-150mm
Mobile phase: Methanol/ H_2O = 50/50
Flow rate: 1.0 ml/min
Temperature: 30°C
Detection: UV254nm

Sample: *o*-Chlorophenol (2.0µg)
m-Chlorophenol (2.0µg)
p-Chlorophenol (4.0µg)



NACALAI TESQUE, INC

AP-1043

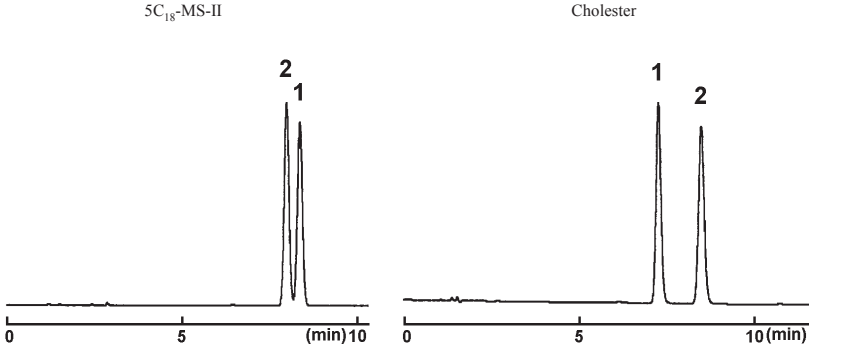
(12) The Others

● Stilbenes

COSMOSIL Application Data

Column:
Column size: 4.6mmI.D.-150mm
Mobile phase: Acetonitrile/ H_2O = 70/30
Flow rate: 1.0 ml/min
Temperature: 30°C
Detection: UV254nm

Sample: 1; *cis*-Stilbene (0.10µg)
2; *trans*-Stilbene (0.20µg)



NACALAI TESQUE, INC

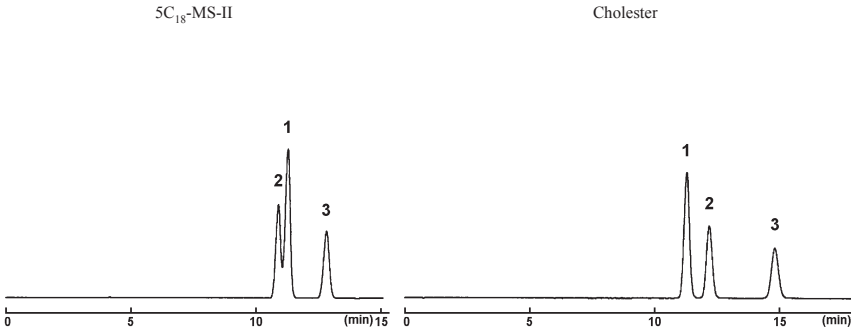
AP-1045

● Halogenated Benzenes

COSMOSIL Application Data

Column:
Column size: 4.6mmI.D.-150mm
Mobile phase: Methanol/ H_2O = 60/40
Flow rate: 1.0 ml/min
Temperature: 30°C
Detection: UV254nm

Sample: 1; Toluene (3.3µg)
2; Chlorobenzene (3.3µg)
3; Bromobenzene (3.3µg)



NACALAI TESQUE, INC

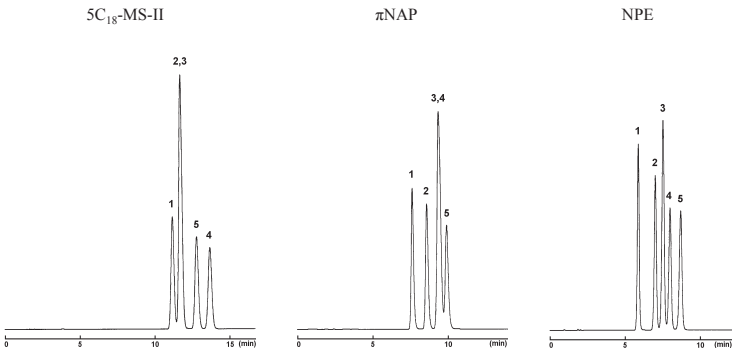
AP-1046

● Fluorinated Benzenes

COSMOSIL Application Data

Column:
Column size: 4.6mmI.D.-150mm
Mobile phase: 5C18-MS-II Methanol/ H_2O = 50/50
πNAP Methanol/ H_2O = 50/50
NPE Methanol/ H_2O = 40/60
Flow rate: 1.0 ml/min
Temperature: 30°C
Detection: UV254nm

Sample: 1; Benzene (6.0µg)
2; Fluorobenzene (2.0µg)
3; *p*-Difluorobenzene (2.0µg)
4; *m*-Difluorobenzene (2.0µg)
5; *o*-Difluorobenzene (2.0µg)



NACALAI TESQUE, INC

AP-1054

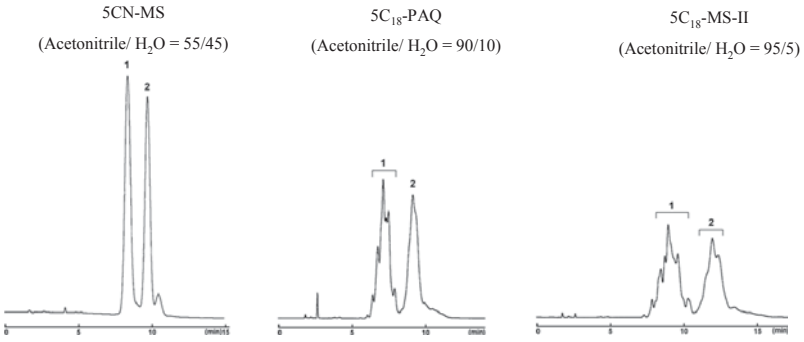
(12) The Others

● Plasticizer (Branched-chain Isomeric Mixture)

COSMOSIL Application Data

Column:
Column size: 4.6mmI.D.-150mm
Mobile phase:
Flow rate: 1.0 ml/min
Temperature: 40°C
Detection: UV220nm

Sample: 1; Diisononyl Phthalate [DINP] (2.0 μg)
2; Diisodecyl Phthalate [DIDP] (2.0 μg)



NACALAI TESQUE, INC

AP-1193

● Plasticizer

COSMOSIL Application Data

Column: 2.5C_{18}-MS-II
Column size: 2.0mmI.D.-100mm
Mobile phase: Methanol/ H_2O = 90/10
Flow rate: 0.35 ml/min
Temperature: 40°C
Detection: UV220nm

Sample:
1; Di-*n*-butyl Phthalate [DBP] (0.05mg/ml)
2; *n*-Butyl Benzyl Phthalate [BBP] (0.05mg/ml)
3; Dioctyl Phthalate [DOP][DEHP] (0.05mg/ml)
4; Di-*n*-octyl Phthalate [DNOP] (0.05mg/ml)
5; Diisononyl Phthalate [DINP] (0.25mg/ml)
6; Diisodecyl Phthalate [DIDP] (0.25mg/ml)
Injection Vol. 5.0μl

NACALAI TESQUE, INC

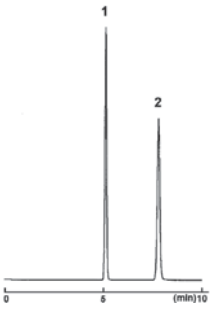
AP-1198

● Uracil and Uridine

COSMOSIL Application Data

Column: HILIC
Column size: 4.6mmI.D.-250mm
Mobile phase: Acetonitrile / H_2O = 90/10
Flow rate: 1.0 ml/min
Temperature: 30°C
Detection: UV254nm

Sample: 1; Uracil (0.1μg)
2; Uridine (0.2μg)



NACALAI TESQUE, INC

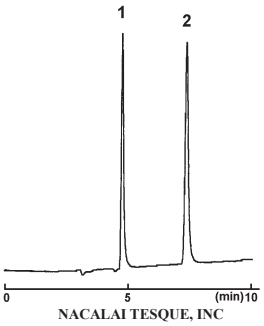
AP-0299

● Glycerol

COSMOSIL Application Data

Column: HILIC
Column size: 4.6mmI.D.-250mm
Mobile phase: Acetonitrile / H_2O = 95/5
Flow rate: 1.0 ml/min
Temperature: 30°C
Detection: RI

Sample: 1; Trimethylene Glycol (20μg)
2; Glycerol (20μg)



NACALAI TESQUE, INC

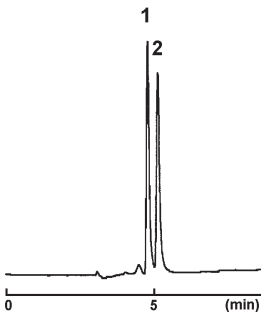
AP-0301

● Ethylene Glycol

COSMOSIL Application Data

Column: HILIC
Column size: 4.6mmI.D.-250mm
Mobile phase: Acetonitrile / H_2O = 95/5
Flow rate: 1.0 ml/min
Temperature: 30°C
Detection: RI

Sample: 1; Trimethylene Glycol (20μg)
2; Ethylene Glycol (20μg)



NACALAI TESQUE, INC

AP-0303

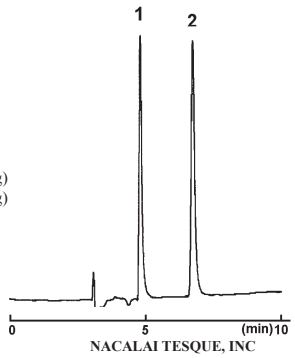
(12) The Others

● Diethylene Glycol

COSMOSIL Application Data

Column: HILIC
Column size: 4.6mmI.D.-250mm
Mobile phase: Acetonitrile / H_2O = 95/5
Flow rate: 1.0 ml/min
Temperature: 30°C
Detection: RI

Sample: 1; Diethylene Glycol (20µg)
2; Glycerol (20µg)



NACALAI TESQUE, INC

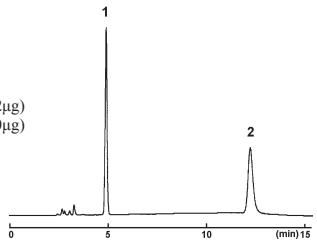
AP-0310

● Oxalic Acid

COSMOSIL Application Data

Column: HILIC
Column size: 4.6mmI.D.-250mm
Mobile phase: Acetonitrile/ 10mmol/l Phosphate Buffer(pH7.0) = 50/50
Flow rate: 1.0 ml/min
Temperature: 30°C
Detection: UV210nm

Sample: 1; Oxamic Acid (0.2µg)
2; Oxalic Acid (1.0µg)



NACALAI TESQUE, INC

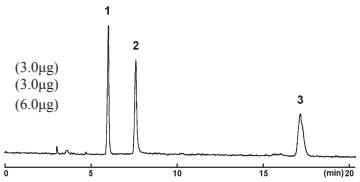
AP-0307

● Hydrophilic Compounds (Ionicity)

COSMOSIL Application Data

Column: HILIC
Column size: 4.6mmI.D.-250mm
Mobile phase: Acetonitrile/ 50mmol/l Ammonium Acetate = 80/20
Flow rate: 1.0 ml/min
Temperature: 30°C
Detection: ELSD

Sample:
1; *meso*-Erythritol〔*meso*-Erythrite〕 (3.0µg)
2; Tris(hydroxymethyl)aminomethane (3.0µg)
3; *DL*-Glyceric Acid (6.0µg)



NACALAI TESQUE, INC

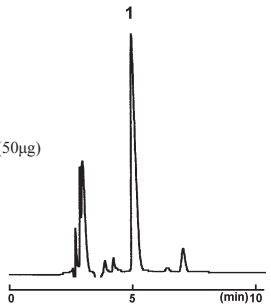
AP-0305

● Fluorine Compounds

COSMOSIL Application Data

Column: $5C_{18}$-AR-II
Column size: 4.6mmI.D.-250mm
Mobile phase: 0.1%TFA-95%Methanol
Flow rate: 1.0 ml/min
Temperature: 30°C
Detection: UV210nm

Sample: Perfluorotetradecanoic Acid (50µg)



NACALAI TESQUE, INC

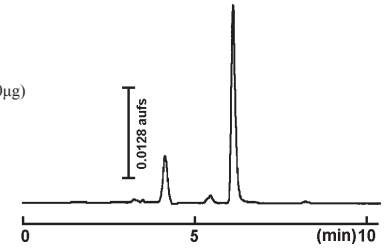
AP-0381

● Urea

COSMOSIL Application Data

Column: HILIC
Column size: 4.6mmI.D.-250mm
Mobile phase: Acetonitrile/H_2O = 90/10
Flow rate: 1.0 ml/min
Temperature: 30°C
Detection: UV210nm

Sample: Urea (20µg)



NACALAI TESQUE, INC

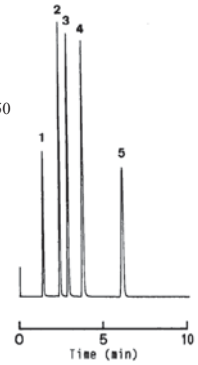
AP-1004

● Acid Compounds

COSMOSIL Application Data

Column: $5C_{18}$-MS-II
Column size: 4.6mmI.D.-150mm
Mobile phase: Acetonitrile / 0.1% Phosphoric Acid = 50/50
Flow rate: 1.0 ml/min
Temperature: 30°C
Detection: UV254nm, 0.32AUFS

Sample: 1; Uracil (0.05µg)
2; Benzoic Acid (1.0µg)
3; *o*-Toluic Acid (1.0µg)
4; *p*-Ethylbenzoic Acid (0.2µg)
5; Benzene



NACALAI TESQUE, INC

AP-0164

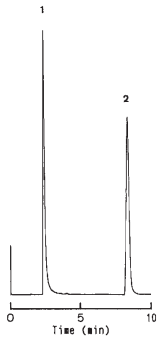
(12) The Others

● Basic Compounds

COSMOSIL Application Data

Column: $5C_{18}$-AR-II
Column size: 4.6mmI.D.-150mm
Mobile phase: Methanol/ 20mmol/l Phosphate Buffer(pH3) = 50/50
Flow rate: 1.0 ml/min
Temperature: 30°C
Detection: UV254nm, 0.2AUFS

Sample: 1; *o*-Ethylpyridine (0.4µg)
2; *N,N*-Dimethylaniline (0.6µg)



NACALAI TESQUE, INC

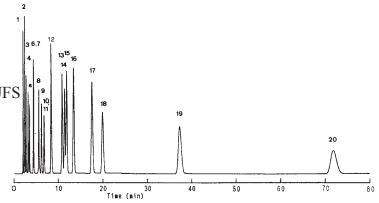
AP-0166

● Monosubstituted Benzenes (20 samples)

COSMOSIL Application Data

Column: $5C_{18}$-MS-II
Column size: 4.6mmI.D.-150mm
Mobile phase: Methanol / H_2O = 60/40
Flow rate: 1.0 ml/min
Temperature: 30°C
Detection: UV254nm, 0.16AUFS
Sample:

1; Benzamide	(0.49µg)		
2; Aniline	(0.4µg)		
3; Phenol	(0.67µg)		
4; Benzonitrile	(0.83µg)		
5; Acetophenone	(0.04µg)		
6; Styrene Oxide	(1.1µg)		
7; Nitrobenzene	(0.06µg)	14; Chlorobenzene	(7.05µg)
8; Methylbenzoate	(0.62µg)	15; Toluene	(5.84µg)
9; Anisole	(0.79µg)	16; Bromobenzene	(15.37µg)
10: Fluorobenzene	(0.65µg)	17; Iodobenzene	(3.66µg)
11; Benzene	(0.79µg)	18; Ethylbenzene	(6.87µg)
12; *N,N*-Dimethylaniline	(0.15µg)	19; *n*-Propylbenzene	(14.1µg)
13; Thioanisole	(0.12µg)	20; *n*-Butylbenzene	(15.93µg)



NACALAI TESQUE, INC

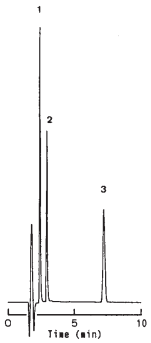
AP-0154

● Furans

COSMOSIL Application Data

Column: $5C_{18}$-MS-II
Column size: 4.6mmI.D.-150mm
Mobile phase: Acetonitrile/ H_2O = 30/70
Flow rate: 1.0 ml/min
Temperature: 30°C
Detection: UV220nm, 0.2AUFS

Sample: 1; Furfuryl alcohol (0.13µg)
2; Furfural (0.25µg)
3; Furan (0.23µg)



NACALAI TESQUE, INC

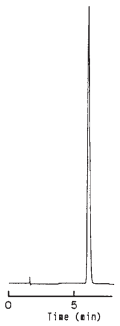
AP-0155

● Phenolphthalein

COSMOSIL Application Data

Column: $5C_{18}$-MS-II
Column size: 4.6mmI.D.-150mm
Mobile phase: Methanol / H_2O = 50/50
Flow rate: 1.0 ml/min
Temperature: 30°C
Detection: UV254nm, 0.1AUFS

Sample: Phenolphthalein (0.6µg)



NACALAI TESQUE, INC

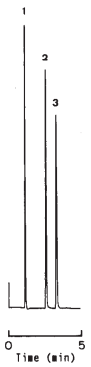
AP-0156

● Anilines

COSMOSIL Application Data

Column: $5C_{18}$-MS-II
Column size: 4.6mmI.D.-150mm
Mobile phase: Acetonitrile / 0.1% Phosphoric Acid = 50/50
Flow rate: 1.0 ml/min
Temperature: 30°C
Detection: UV254nm

Sample: 1; Aniline
2; *p*-Nitroaniline
3; *2,4*-Dinitroaniline



NACALAI TESQUE, INC

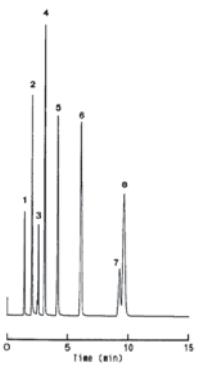
AP-0160

● Esters and others

COSMOSIL Application Data

Column: $5C_{18}$-MS-II
Column size: 4.6mmI.D.-150mm
Mobile phase: Acetonitrile/ H_2O = 50/50
Flow rate: 1.0 ml/min
Temperature: 30°C
Detection: UV254nm, 0.32AUFS

Sample: 1; Uracil
2; Pyridine
3; Phenol
4; Ethyl *p*-Hydroxybenzoate
5; *n*-Propyl *p*-Hydroxybenzoate
6; Methyl Salicylate
7; Toluene
8; Ethyl Salicylate



NACALAI TESQUE, INC

AP-0163

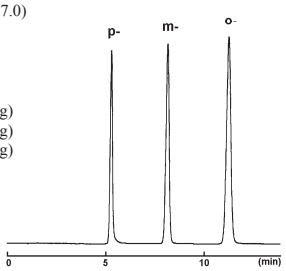
(12) The Others

● Aminophenols

COSMOSIL Application Data

Column: $5C_{18}$-PAQ
Column size: 4.6mmI.D.-250mm
Mobile phase: 20mmol/l Phosphate buffer(pH7.0)
Flow rate: 1.0 ml/min
Temperature: 30°C
Detection: UV254nm

Sample: *p*-Aminophenol (0.33µg)
m-Aminophenol (1.00µg)
o-Aminophenol (1.67µg)



NACALAI TESQUE, INC

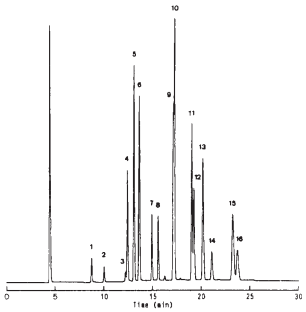
AP-1059

● Polyaromatic Compounds

COSMOSIL Application Data

Column: $5C_{18}$-AR-II
Column size: 4.6mmI.D.-150mm
Mobile phase: A; Methanol / H_2O = 70/30
B; Methanol
B conc.0→100%
3→15min Linear gradient
Flow rate: 1.0 ml/min
Temperature: 30°C
Detection: UV254nm, 0.64AUFS

Sample:
1; Naphthalene
2; Acenaphthylene
3; Acenaphthene
4; Fluorene
5; Phenanthrene
6; Anthracene
7; Fluoranthene
8; Pyrene
9; Benz[*a*]anthracene
10; Chrysene
11; Benzo[*b*]fluoranthene
12; Benz[*a*]pyrene
13; Benzo[*a*]pyrene
14; *1,2:5,6*-Dibenzanthracene
15; Benzo[*g,h,i*]perylene
16; Indeno[*1,2,3-c,d*]pyrene



NACALAI TESQUE, INC

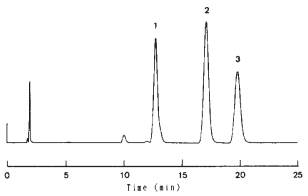
AP-0161

● Benzylpyridines

COSMOSIL Application Data

Column: 5PYE
Column size: 4.6mmI.D.-150mm
Mobile phase: Methanol/ 20mmol/l KH_2PO_4 = 60/40
Flow rate: 1.0 ml/min
Temperature: 30°C
Detection: UV254nm

Sample: 1; *2*-Benzylpyridine
2; *3*-Benzylpyridine
3; *4*-Benzylpyridine



NACALAI TESQUE, INC

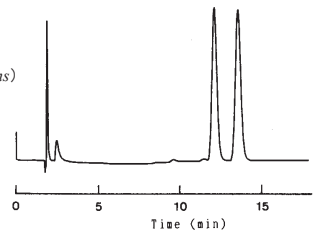
AP-0182

● Carvylacetate

COSMOSIL Application Data

Column: 5PYE
Column size: 4.6mmI.D.-150mm
Mobile phase: Methanol / H_2O = 70/30
Flow rate: 1.0 ml/min
Temperature: 30°C
Detection: UV215nm

Sample: Carvylacetate(*cis*, *trans*)



NACALAI TESQUE, INC

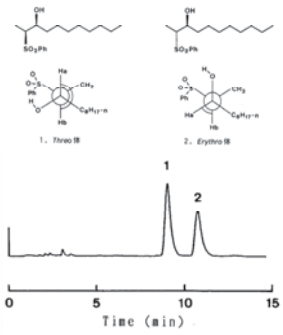
AP-0184

● Diastereomers

COSMOSIL Application Data

Column: 5PYE
Column size: 4.6mmI.D.-150mm
Mobile phase: Methanol / H_2O = 80/20
Flow rate: 1.0 ml/min
Temperature: 30°C
Detection: UV254nm

Sample: 1; *Threo* form
2; *Erythro* form



NACALAI TESQUE, INC

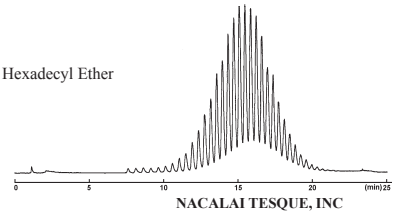
AP-0186

● Surfactant

COSMOSIL Application Data

Column: 5PBB-R
Column size: 4.6mmI.D.-150mm
Mobile phase: A: Methanol/ H_2O = 80/20
B: Tetrahydrofuran/ Methanol = 30/70
B conc. 30→100% 20min Linear gradient
Flow rate: 1.0 ml/min
Temperature: 30°C
Detection: ELSD

Sample: Polyoxyethylene Hexadecyl Ether
Sample conc. 10mg/ml
Inj.Vol. 5.0µl



NACALAI TESQUE, INC

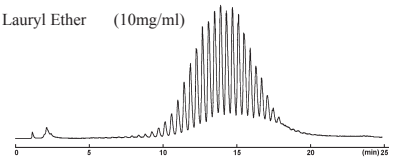
AP-1202

(12) The Others

● Surfactant

COSMOSIL Application Data

Column: 5PBB-R
Column size: 4.6mmI.D.-150mm
Mobile phase: A: Methanol/ H_2O = 80/20
B: Tetrahydrofuran/ Methanol = 30/70
B conc. 30→100% 20min Linear gradient
Flow rate: 1.0 ml/min
Temperature: 30°C
Detection: ELSD
Sample: Polyoxyethylene Lauryl Ether (10mg/ml)
Inj.Vol. 5.0µl



NACALAI TESQUE, INC

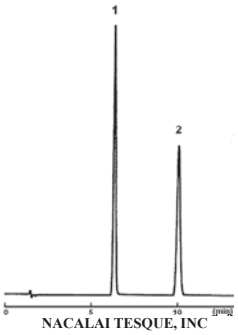
AP-1201

● Aromatic Compounds

COSMOSIL Application Data

Column: 5PBB-R
Column size: 4.6mmI.D.-150mm
Mobile phase: Acetonitrile/H_2O = 70/30
Flow rate: 1.0 ml/min
Temperature: 30°C
Detection: UV254nm

Sample: 1; Diphenylmethane (5.80µg)
2; Fluorene (0.13µg)



NACALAI TESQUE, INC

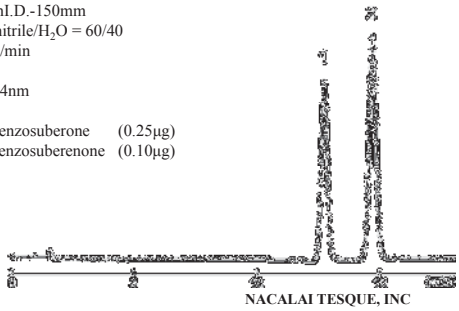
AP-0358

● Aromatic Compounds

COSMOSIL Application Data

Column: 5PBB-R
Column size: 4.6mmI.D.-150mm
Mobile phase: Acetonitrile/H_2O = 60/40
Flow rate: 1.0 ml/min
Temperature: 30°C
Detection: UV254nm

Sample: 1; Dibenzosuberone (0.25µg)
2; Dibenzosuberenone (0.10µg)



NACALAI TESQUE, INC

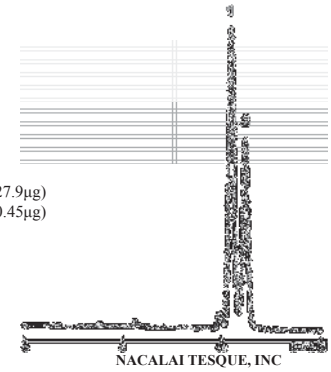
AP-0360

● Aromatic Compounds

COSMOSIL Application Data

Column: 5PBB-R
Column size: 4.6mmI.D.-150mm
Mobile phase: Methanol/H_2O = 70/30
Flow rate: 1.0 ml/min
Temperature: 30°C
Detection: UV254nm

Sample: 1; *n*-Propylbenzene (27.9µg)
2; Allylbenzene (0.45µg)



NACALAI TESQUE, INC

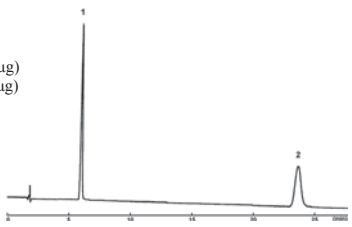
AP-0362

● Aromatic Compounds

COSMOSIL Application Data

Column: 5PBB-R
Column size: 4.6mmI.D.-150mm
Mobile phase: Methanol/H_2O = 90/10
Flow rate: 1.0 ml/min
Temperature: 30°C
Detection: UV254nm

Sample: 1; *1,1'*-Binapthyl (0.60µg)
2; Perylene (0.72µg)



NACALAI TESQUE, INC

AP-0356

● Coloring Agent (CBB)

COSMOSIL Application Data

Column: $5C_{18}$-MS-II
Column size: 4.6mmI.D.-150mm
Mobile phase: Methanol/ 20mmol/l Phosphate buffer(pH2.5) = 70/30
Flow rate: 1.0 ml/min
Temperature: 30°C
Detection: UV254nm

Sample: Coomassie Brilliant Blue G-250 (3.0µg)

NACALAI TESQUE, INC

AP-1061

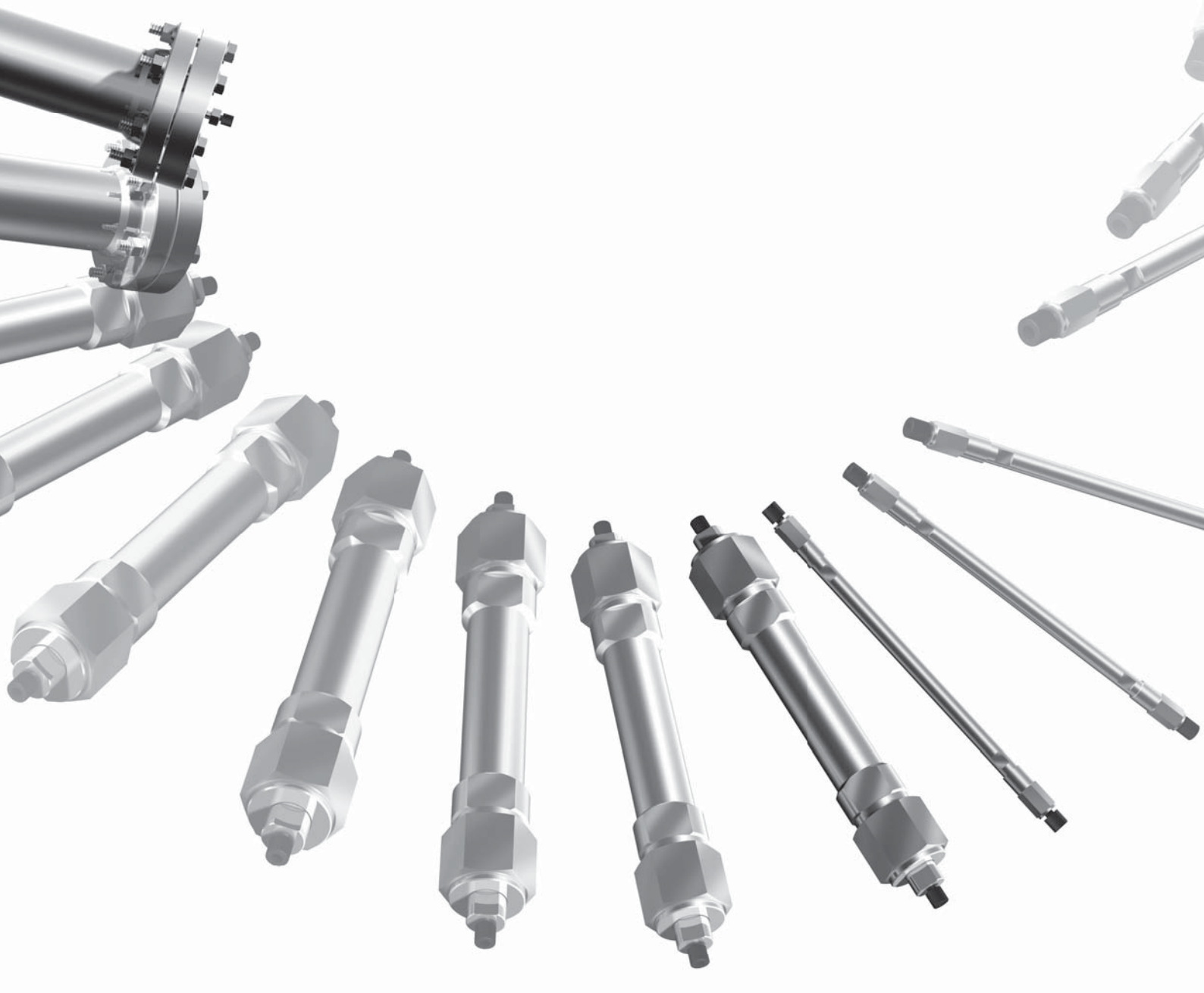
4. Reference List

COSMOSIL HPLC columns have been referenced in more than 5000 scientific literatures. The followings are recent references of COSMOSIL special columns. Copies are not available directly from us due to the copyright.

No.	Column	TITLE	AUTHOR	JOURNAL	YEAR	VOL. (ISSUE)	PAGE
1	Cholester πNAP	Four new glucosides from the aerial parts of Mediasia macrophylla	Shin-ichiro Kurimoto, Mamoru Okasaka, Yoshiki Kashiwada, Olimjon K. Kodzhimatov and Yoshihisa Takaishi	Journal of Natural Medicines	2011	65 (1)	180-185
2	Cholester	Inhibitory Effect of Cyclic Trihydroxamate Siderophore, Desferrioxamine E, on the Biofilm Formation of Mycobacterium Species	Shunsuke Ishida, Masayoshi Arai, Hiroki Niikawa and Motomasa Kobayashi	Biological & Pharmaceutical Bulletin	2011	34 6)	917-920
3	Cholester	A study on thermal stability of lycopene in tomato in water and oil food systems using response surface methodology	Honest H. Kessy, Huanwei Zhang, Lianfu Zhang	International Journal of Food Science & Technology	2011	46 (1)	209-215
4	Cholester	Melicodenines A and B, novel Diels–Alder type adducts isolated from Melicope denhamii	Ken-ichi Nakashima, Masayoshi Oyama, Tetsuro Ito, Joko Ridho Witono, Dedy Darnaedi, Toshiyuki Tanaka, Jin Murata, Munekazu Iinuma	Tetrahedron Letters	2011	52 (36)	4694-4696
5	Cholester	Effects of a Citrus depressa Hayata (shiikuwasa) extract on obesity in high-fat diet-induced obese mice	Young-Sil Lee, Byung-Yoon Cha, Kiyoto Saito, Sun-Sil Choi, Xiao Xing Wang, Bong-Keun Choi, Takayuki Yonezawa, Toshiaki Teruya, Kazuo Nagai, Je-Tae Woo	Phytomedicine	2011	18 (8-9)	648-654
6	5C18-MS-II Cholester πNAP	Triterpenes and a triterpene glucoside from Dysoxylum cumingianum	Shin-ichiro Kurimoto, Yoshiki Kashiwada, Kuo-Hsiung Lee, Yoshihisa Takaishi	Phytochemistry	2011	In Press	
7	Cholester	Direct Dehydrative Pyridylthio-Glycosidation of Unprotected Sugars in Aqueous Media Using *2*-Chloro-*1,3*-dimethylimidazolinium Chloride as a Condensing Agent	Naoki Yoshida, Dr. Masato Noguchi, Dr. Tomonari Tanaka, Takeshi Matsumoto, Naoya Aida, Dr. Masaki Ishihara, Dr. Atsushi Kobayashi, Prof. Dr. Shin-ichiro Shoda	Chemistry – An Asian Journal	2011	6 (7)	1876-1885
8	Cholester	Seven new dammarane triterpenes from the floral spikes of Betula platyphylla var. japonica	Juan Xiong, Masatoshi Taniguchi, Yoshiki Kashiwada, Takashi Yamagishi and Yoshihisa Takaishi	Journal of Natural Medicines	2011	65 (1)	217–223
9	Cholester	Osteoclast-forming suppressing compounds, gargalols A, B, and C, from the edible mushroom Grifola gargal	Jing Wu, Jae-Hoon Choi, Miyuki Yoshida, Hirofumi Hirai, Etsuko Harada, Kikuko Masuda, Tomoyuki Koyama, Kazunaga Yazawa, Keiichi Noguchi, Kazuo Nagasawa	Tetrahedron	2011	67 (35)	6576-6581
10	Cholester	Contribution of cinnamic acid analogues in rosmarinic acid to inhibition of snake venom induced hemorrhage	Hnin Thanda Aung, Tadashi Furukawa, Toshiaki Nikai, Masatake Niwa, Yoshiaki Takaya	Bioorganic & Medicinal Chemistry	2011	19 (7)	2392-2396
11	Cholester	Applanatines A–E from the culture broth of Ganoderma applanatum	Keiji Fushimi, Madoka Horikawa, Kaori Suzuki, Atsushi Sekiya, Susumu Kanno, Susumu Shimura, Hirokazu Kawagishi	Tetrahedron	2010	66 (48)	9332-9335
12	Cholester 140C18 PREP	Four New Cembrane Diterpenes Isolated from an okinawan Soft Coral Lobophytum crassum with Inhibitory Effects on Nitric Oxide Production	Mpanzu Wanzola, Takaaki Furuta, Yasuhisa Kohno, Shunichi Fukumitsu, Shuhei Yasukochi, Kosuke Watari, Chiaki Tanaka, Ryuichi Higuchi and Tomofumi Miyamoto	CHEMICAL & PHARMACEUTICAL BULLETIN	2010	58 (9)	1203-1209
13	Cholester	Nobiletin improves hyperglycemia and insulin resistance in obese diabetic ob/ob mice	Young-Sil Lee, Byung-Yoon Cha, Kiyoto Saito, Hiroshi Yamakawa, Sun-Sil Choi, Kohji Yamaguchi, Takayuki Yonezawa, Toshiaki Teruya, Kazuo Nagai, Je-Tae Woo	Biochemical Pharmacology	2010	79 (11)	1674-1683
14	Cholester	Study of solvation processes on cholesterol bonded phases	Boguslaw Buszewski, Szymon Bocian, Maria Matyska, Joseph Pesek	Journal of Chromatography A	2010	1218 (3)	441-448
15	Cholester	Study of the retention and selectivity of cholesterol bonded phases with different linkage spacers	Szymon Bocian, Maria Matyska, Joseph Pesek, Boguslaw Buszewski	Journal of Chromatography A	2010	1217 (44)	6891-6897
16	Cholester 5C18-AR-II	Isolation and structure of koshikalide, a *14*-membered macrolide from the marine cyanobacterium Lyngbya sp.	Arihiro Iwasaki, Toshiaki Teruya and Kiyotake Suenaga	Tetrahedron Letters	2010	51 (6)	959-960
17	Cholester πNAP	A C14-polyacetylenic glucoside with an α-pyrone moiety and four C10-polyacetylenic glucosides from Mediasia macrophylla	Shin-ichiro Kurimoto, Mamoru Okasaka, Yoshiki Kashiwada, Olimjon K. Kodzhimatov and Yoshihisa Takaishi	Phytochemistry	2010	71 (5-6)	688-692
18	πNAP	Development and validation of a stability-indicating LC method for determining Palonosetron hydrochloride, its related compounds and degradation products using naphthalethyl stationary phase	M. Vishnu Murthy, Katkam Srinivas, Ramesh Kumar, K. Mukkanti	Journal of Pharmaceutical and Biomedical Analysis	2011	56 (2)	429-435
19	πNAP	Maklamicin, an Antibacterial Polyketide from an Endophytic Micromonospora sp.	Yasuhiro Igarashi, Hiromu Ogura, Kazuo Furihata, Naoya Oku, Chantra Indananda, and Arinthip Thamchaipenet	J. Nat. Prod.	2011	74 (4)	670–674
20	πNAP	Gneyulins A and B, Stilbene Trimers, and Noidesols A and B, Dihydroflavonol-C-Glucosides, from the Bark of Gnetum gnemonoides	Yoko Shimokawa, Yusuke Akao, Yusuke Hirasawa, Khalijah Awang, A. Hamid A. Hadi, Seizo Sato, Chihiro Aoyama, Jiro Takeo, Motoo Shiro and Hiroshi Morita	J. Nat. Prod.	2010	73 (4)	763–767
21	HILIC	Stationary and mobile phases in hydrophilic interaction chromatography: a review	Pavel Jandera	Analytica Chimica Acta	2011	692 (1-2)	1-25
22	HILIC Sugar-D	Chromatographic characterization of hydrophilic interaction liquid chromatography stationary phases: Hydrophilicity, charge effects, structural selectivity, and separation efficiency	Yuusuke Kawachi, Tohru Ikegami, Hirotaka Takubo, Yuka Ikegami, Masatoshi Miyamoto, Nobuo Tanaka	Journal of Chromatography A	2011	1218 (35)	5903-5919

No.	Column	TITLE	AUTHOR	JOURNAL	YEAR	VOL. (ISSUE)	PAGE
23	HILIC	Retention and selectivity of stationary phases for hydrophilic interaction chromatography	Yong Guo, Sheetal Gaiki	Journal of Chromatography A	2011	1218 (35)	5920–5938
24	HILIC	The different decomposition properties of diazolidinyl urea in cosmetics and patch test materials	Takahiro Doi, Keiji Kajimura, Shuzo Taguchi	Contact Dermatitis	2011	65 (2)	81–91
25	HILIC	A Novel Glucosylation Reaction on Anthocyanins Catalyzed by Acyl–Glucose–Dependent Glucosyltransferase in the Petals of Carnation and Delphinium	Yuki Matsuba, Nobuhiro Sasaki, Masayuki Tera, Masachika Okamura, Yutaka Abe, Emi Okamoto, Haruka Nakamura, Hisakage Funabashi, Makoto Takatsu, Mikako Saito, Hideaki Matsuoka, Kazuo Nagasawa and Yoshihiro Ozeki	The Plant Cell	2010	22 (10)	3374–3389
26	HILIC	Molecular identification of unsaturated uronate reductase prerequisite for alginate metabolism in Sphingomonas sp. A1	Ryuichi Takase, Akihito Ochiai, Bunzo Mikami, Wataru Hashimoto, Kousaku Murata	Biochimica et Biophysica Acta (BBA) – Proteins & Proteomics	2010	1804 (9)	1925–1936
27	HILIC	Hepatoprotective Effects of Flavonoids from Shekwasha (Citrus depressa) against *D*-Galactosamine-Induced Liver Injury in Rats	Toshiyuki AKACHI, Yasuyuki SHIINA, Yayoi OHISHI, Takumi KAWAGUCHI, Hirokazu KAWAGISHI, Tatsuya MORITA, Makoto MORI and Kimio SUGIYAMA	Journal of Nutritional Science and Vitaminology	2010	56 (1)	60–67
28	HILIC	Determination of isoascorbic acid in fish tissue by hydrophilic interaction liquid chromatography–ultraviolet detection	Spyros Drivelos, Marilena E. Dasenaki and Nikolaos S. Thomaidis	Analytical and Bioanalytical Chemistry	2010	397 (6)	2199–2210
29	HILIC	Approach to hydrophilic interaction chromatography column selection: Application to neurotransmitters analysis	Raluca-Ioana Chirita, Caroline West, Adriana-Luminita Finaru, Claire Elfakir	Journal of Chromatography A	2010	1217 (18)	3091–3104
30	HILIC $5C_{18}$-MS-II	Inhibitory Effects of Acylated Acyclic Sesquiterpene Oligoglycosides from the Pericarps of Sapindus rarak on Tumor Necrosis Factor-α-Induced Cytotoxicity	Toshio Morikawa, Yuanyuan Xie, Kiyofumi Ninomiya, Masaki Okamoto, Osamu Muraoka, Dan Yuan, Masayuki Yoshikawa and Takao Hayakawa	CHEMICAL & PHARMACEUTICAL BULLETIN	2010	58 (9)	1276–1280
31	HILIC	Unusual amino acid derivatives from the mushroom Pleurocybella porrigens	Takumi Kawaguchi, Tomohiro Suzuki, Yuka Kobayashi, Shinya Kodani, Hirofumi Hirai, Kaoru Nagai and Hirokazu Kawagishi	Tetrahedron	2010	66 (2)	504–507
32	Sugar-D	DedA Protein Relates to Action-Mechanism of Halicyclamine A, a Marine Spongean Macrocyclic Alkaloid, as an Anti-dormant Mycobacterial Substance	Masayoshi Arai, Liu Liu, Takao Fujimoto, Andi Setiawan and Motomasa Kobayashi	Marine Drugs	2011	9 (6)	984–993
33	Sugar-D	Efficient ^{1}H-NMR Quantitation and Investigation of *N*-Acetyl-*D*-glucosamine (GlcNAc) and N,N'-Diacetylchitobiose $(GlcNAc)_2$ from Chitin	Fu-Chien Liu, Chung-Ren Su, Tzi-Yi Wu, Shyh-Gang Su, Huey-Lang Yang, John Han-You Lin and Tian-Shung Wu	Int. J. Mol. Sci.	2011	12	5828–5843
34	Sugar-D	New radiosynthesis of 2-deoxy-2-[^{18}F]fluoroacetamido-d-glucopyranose and its evaluation as a bacterial infections imaging agent	Miguel E. Martínez, Yasushi Kiyono, Sakon Noriki, Kunihiro Inai, Katheryn S. Mandap, Masato Kobayashi, Tetsuya Mori, Yuji Tokunaga, Vijay N. Tiwari, Hidehiko Okazawa, Yasuhisa Fujibayashi, Tatsuo Ido	Nuclear Medicine and Biology	2011	38 (6)	807–817
35	Sugar-D	Molecular Cloning and Characterization of a β-l-Arabinobiosidase in Bifidobacterium longum That Belongs to a Novel Glycoside Hydrolase Family	Kiyotaka Fujita, Shiho Sakamoto, Yuki Ono, Masahiro Wakao, Yasuo Suda, Kanefumi Kitahara and Toshihiko Suganuma	The Journal of Biological Chemistry	2011	286	5143–5150
36	Sugar-D	New Phenylpropanoid Glycosides from Juniperus communis var. depressa	Naoki IIDA, Yuka INATOMI, Hiroko MURATA, Jin MURATA, Frank A. LANG, Toshiyuki TANAKA, Tsutomu NAKANISHI and Akira INADA	CHEMICAL & PHARMACEUTICAL BULLETIN	2010	58 (5)	742–746
37	Sugar-D	Novel phenolic glycosides, adenophorasides A–E, from Adenophora roots	Yuka Koike, Motonori Fukumura, Yasuaki Hirai, Yumiko Hori, Shiho Usui, Toshiyuki Atsumi and Kazuo Toriizuka	Journal of Natural Medicines	2010	64(3)	245–251
38	Sugar-D	Quantitative determination of potent α-glucosidase inhibitors, salacinol and kotalanol, in Salacia species using liquid chromatography–mass spectrometry	Osamu Muraoka, Toshio Morikawa, Sohachiro Miyake, Junji Akaki, Kiyofumi Ninomiya and Masayuki Yoshikawa	Journal of Pharmaceutical and Biomedical Analysis	2010	52 (5)	770–773
39	Sugar-D	Structural characterization of N-linked oligosaccharides of Defibrase from Agikistrodon acutus by sequential exoglycosidase digestion and MALDI-TOF mass spectrometry	Xiaoqing Luo, Huaxin Yang, Chenggang Liang and Shaohong Jin	Toxicon	2010	55 (2–3)	421–429
40	CNT	Thin-Film Transistors with Length-Sorted DNA-Wrapped Single-Wall Carbon Nanotubes	Yuki Asada, Yasumitsu Miyata, Kazunari Shiozawa, Yutaka Ohno, Ryo Kitaura, Takashi Mizutani, and Hisanori Shinohara	J. Phys. Chem. C	2011	115 (1)	270–273
41	CNT-300 CNT-1000 CNT-2000	Chromatographic Separation of Highly Soluble Diamond Nanoparticles Prepared by Polyglycerol Grafting	Li Zhao, Tatsuya Takimoto, Masaaki Ito, Naoko Kitagawa, Takahide Kimura, Naoki Komatsu	Angewandte Chemie	2011	123 (6)	1424–1428
42	CNT-300 CNT-1000 CNT-2000	High-Performance Thin-Film Transistors with DNA-Assisted Solution Processing of Isolated Single-Walled Carbon Nanotubes	Yuki Asada, Yasumitsu Miyata, Yutaka Ohno, Ryo Kitaura, Toshiki Sugai, Takashi Mizutani, Hisanori Shinohara	Advanced Materials	2010	22 (24)	2698–2701
43	CNT-300 CNT-1000 CNT-2000	Chromatographic Length-Separation and Photoluminescence Study on DNA-Wrapped Single-Wall and Double-Wall Carbon Nanotubes	Yuki Asada, Toshiki Sugai, Ryo Kitaura and Hisanori Shinohara	Journal of nanomaterials	2009	2009	8

VI Technical Notes



1.FAQ

(1) FAQ and Troubleshooting List

(2) FAQ

Q1. What is the pressure limit of column?

Column		Pressure Limit
UHPLC Column	COSMOSIL 2.5C_{18}-MS-II COSMOSIL 2.5Cholester COSMOSIL 2.5πNAP	30 MPa
CNT Column	COSMOSIL CNT Series	15 MPa or less
Other Column	Analysis Column (I.D.1.0–7.5 mm)	20 MPa or less
	Analysis Column (I.D.10.0 mm or more)	15 MPa or less

Attention;
A large pressure change may deteriorate columns even within the recommended pressure range.

Q2. What is the flow rate limit?

You can raise the flow rate under the pressure limits stated on Q1

Attention;
We recommend using standard flow rate described in the technical information 2 on page 189. Generally, higher column pressure corresponds to shorter column lifetime.

Q3. What is the recommended pH range?

Column		Recommended pH Range
COSMOSIL (silica gel base) Series	COSMOSIL C_{18}-MS-II	pH 2–10
	COSMOSIL C_{18}-AR-II	pH 1.5–7.5
	Other Columns	pH 2–7.5
COSMOGEL (polymer base) Series	COSMOGEL IEX Series	pH 2–12

Attention;
Table above shows tolerant range for the packing material. Adjust the pH for ionic samples.

Q4. What is the concentration of buffer and salt?

Column		Buffer and Salt Concentration
Reversed Phase, Normal Phase, HILIC		Buffer concentration: 0.005–0.1 mol/l Additive concentration (trifluoroacetic acid, formic acid or acetic acid)：0.1–1.0%
Ion Exchange	COSMOGEL IEX Series	Buffer concentration: 0.02–0.1 mol/l Water miscible organic solvent concentration limit (e.g., methanol): 20% or less
Gel Filtration	COSMOSIL Diol Series	Buffer concentration limit: 0.5 mol/l or less Salt concentration limit: 0.5 mmol/l or less
Hydrophobic Interaction	COSMOSIL HIC	Buffer concentration limit: 0.5 mol/l or less Salt concentration limit: 2 mmol/l or less

Attention;
1. Insoluble compounds may clog columns. Filter buffers or salt solution before using.
2. Deposition of salt during analysis may deteriorate columns or equipments. Use the column under the concentration which salt does not precipitate.
3. Salt often precipitates when the organic solvent is mixed with the solution. Be careful when mixing mobile phases.
4. When equipment or a column contains organic solvent, replace mobile phase with salt-free mobile phase first before using salt-containing mobile phase.

Q5. How do I adjust mobile phase?

Please refer to Technical Information 1 on page 187.

Attention;

1. Adjust mobile phase ratio and buffer concentration exactly each time because concentration and pH may affect separation performance.
2. Degas the solvent after mixing.

Q6. What solvent grade should I use for the mobile phase?

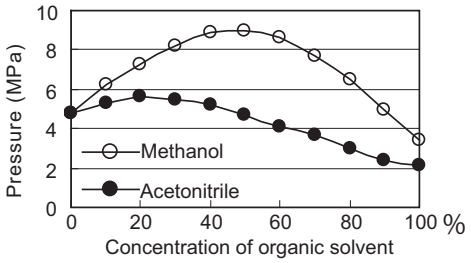
We recommend using HPLC grade solvents, please refer to page 76.

Attention;

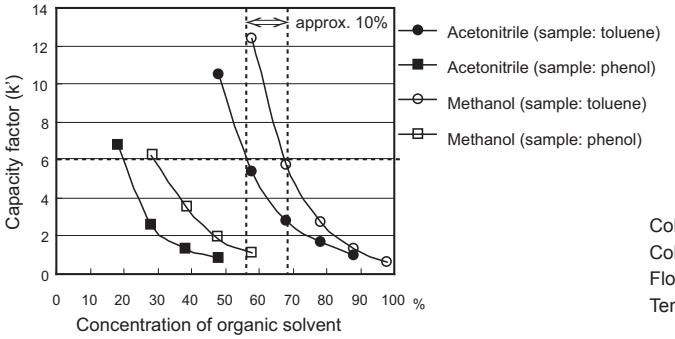
GR grade solvents are not suitable for gradient analysis or micro-scale analysis because they contain impurities that have ultraviolet adsorption causing unstable baseline or inaccurate detection. This is especially problematic in short wavelength (210–220nm). Antioxidant containing GR grade solvents (e.g.. tetrahydrofuran, chloroform) may produce ghost peaks. GR grade trifluoroacetic acid may be chemically unstable and not recommended for HPLC.

Q7. What is the difference between acetonitrile and methanol?

Table below shows difference between acetonitrile and methanol

<table>
<tr><th></th><th>Acetonitrile (for HPLC)</th><th>Methanol (for HPLC)</th></tr>
<tr><td>Pressure</td><td colspan="2"> 

Column 5C18-MS-II
Column size 4.6 mm I.D. x 150 mm
Flow rate 1.0ml/min
Temperature 30°C

Back pressure differs depending on species of organic solvents and mixing ratio. Back pressure of acetonitrile is lower than that of methanol at the same concentration.</td></tr>
<tr><td>Elution Strength</td><td colspan="2"> 

Column 5C18-MS-II
Column size 4.6mmI.D.-150mm
Flow rate 1.0ml/min
Temperature 30°C

Elution strength of acetonitrile/water is stronger than methanol/ water at the same concentration. Within the range of about 30–80% in the concentration of the organic solvent, elution strength of acetonitrile/water is almost equivalent to that of methanol/water which was raised methanol concentration about 10% (e.g., acetonitrile : water = 60 : 40 → methanol : water = 70 : 30)</td></tr>
<tr><td>Absorbance</td><td>Acetonitrile has a lower UV absorbance in far UV region (less than 250 nm) .</td><td>Methanol has a higher UV absorbance than acetonitrile in far UV region (less than 250 nm).</td></tr>
<tr><td>Degas of Mobile Phase</td><td>When acetonitrile is mixed with water, it is endothermic. It is difficult to degas.</td><td>When methanol is mixed with water, it is exothermic. It is easy to degas.</td></tr>
</table>

Q8. Which mobile phase can be used for LC/MS or ELSD detector?

Volatile solvent should be used for LC/MS detector or ELSD. Phosphoric acid buffer can not be used.

Regent	Usable solvent / Additive
Solvent	Methanol, ethanol, acetonitrile, water and others
pH Adjusting Reagent	Acetic acid, formic acid, trifluoroacetic acid, ammonia water, ammonium acetate, ammonium formate and others
Ion Pair Reagent	Dibutylamine, triethylamine and others

Attention;
A small amount of non-volatile solvent, such as DMSO (dimethylsulfoxide) or DMF (dimethylformamide) can be used if they are mixed with methanol or acetonitrile. However, if the concentration becomes higher, detection sensitivity may decrease.

Q9. What should I pay attention to when I use ion-pairing reagents?

- Concentration of ion–pairing reagent should be 5–10 mmol/l.
- Use mobile phase of pH7 for acidic ion–pairing reagents and pH2.5 for basic ion–pairing reagents.
- Have enough equilibration time.
- Please refer to page 78 for choosing conditions.

Attention;

1. Higher ion pair concentration will result longer retention time.
2. Adjust pH of mobile phase so that sample is well ionized.
3. Need longer equilibration time compared to mobile phase without ion–pairing reagent.
4. Use a column exclusively with ion–pairing reagents since it is difficult to eliminate it from the column.

Q10. What flow direction should I use for the mobile phase?

Pump mobile phase through the direction specified on the column label.

Attention;

Pumping mobile phase in reverse direction may deteriorate packing material and decrease theoretical plates. Furthermore, impurities previously adsorbed on the column tip may loosen, causing contaminated detector, tubing, and noise.

Q11. What is the recommended temperature of columns?

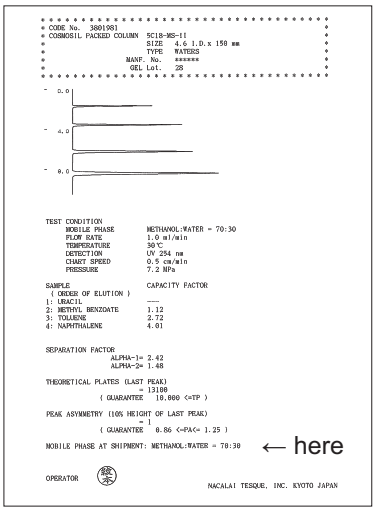
The maximum temperature is 60°C, but the recommended temperaturerange is 20–50°C.

Attention;

1. Operating analysis at high temperature under alkaline or acidic condition may shorten column lifetime.
2. Maintain constant temperature during analysis as column temperature affects retention time. Generally, higher temperature lowers column pressure and shortens retention time.

Q12. What is the shipping solvent?

It depends on the column type. Please refer to the certificate of analysis inside the column box.


CODE No. 380198
COSMOSIL PACKED COLUMN 5C18-MS-II
SIZE 4.6 I.D. x 150 mm
TYPE WATERS
MANF. No. ******
GEL Lot. 28

TEST CONDITION
MOBILE PHASE METHANOL:WATER = 70:30
FLOW RATE 1.0 ml/min
TEMPERATURE 30℃
DETECTION UV 254 nm
CHART SPEED 0.5 cm/min
PRESSURE 7.2 MPa

SAMPLE (ORDER OF ELUTION) CAPACITY FACTOR
1: URACIL ---
2: METHYL BENZOATE 1.12
3: TOLUENE 2.72
4: NAPHTHALENE 4.01

SEPARATION FACTOR
ALPHA-1= 2.42
ALPHA-2= 1.48

THEORETICAL PLATES (LAST PEAK)
= 13100
(GUARANTEE 10,000 <=TP)

PEAK ASYMMETRY (10% HEIGHT OF LAST PEAK)
= 1
(GUARANTEE 0.86 <=PAC= 1.25)

MOBILE PHASE AT SHIPMENT: METHANOL:WATER = 70:30 ← here

OPERATOR

NACALAI TESQUE, INC. KYOTO JAPAN

Q13. How do I wash columns?

1. How to eliminate buffer, salt or acid from a column?
   Wash the column with solvent without buffer, salt or acid for 10–15 min. and store.
   (E.g., When methanol : 20 mmol/l phosphoric acid buffer = 50 : 50 is used, wash with methanol : water = 50 : 50)
   Attention;
   If ratio of organic solvent and water is changed, salt may precipitate.

2. How to wash impurities in a column to achieve a stable base line?
   Dissolve sample well, and use solvent with strong elution properties.

Type	Usable Solvent / Additive
Reversed Phase	(1) Sample is not protein Methanol or tetrahydrofuran Cleaning Solution Kit for Reversed Phase HPLC Columns (Product No. 08966–30) (2) Sample is protein 50–70% acetonitrile : water containing 0.1% trifluoroacetic acid. Attention; Some proteins may precipitate when concentration of organic solvent is high.
Normal Phase	Methanol, tetrahydrofuran, ethanol
Column for Fullerene Separation	*1,2,4*–trichlorobenzene and others
Sugar–D, NH_2, HILIC	Acetonitrile : water = 50 : 50 *Sugar–D and HILIC can be washed with 100% water.

Attention;
Do not use alkali solution (pH 7.5 or higher), which dissolve silica gel. Do not use strong acid solution (pH 1.5 or less) that may cleave bonded stationary phase.

3. How to eliminate impurities clogging the column entry end?
   Pumping solvent through in a reversed direction at half the speed of ordinal analysis.
   Attention;
   Disconnect column from the detector during washing.
4. If column pressure is high, please refer to Technical Information 3 on page 191.

Q14. How do I store columns?

1. Store for short time (a few days)
   Wash the column with solvent without buffer, salt or acid for 10–15 min. and store
   (E.g., When methanol : 20 mmol/l phosphoric acid buffer = 50 : 50 is used, wash with methanol : water = 50 : 50)
2. Store for long time (one month or more)
   Replace solvent with the following solvent to avoid fungus, dry and deterioration of column.

Column	Concentration of Buffer or Base
Reversed Phase	Storage Solution for Reversed Phase HPLC Columns (Product No. 08967–20) Methanol : water = 70 : 30, Acetonitrile : water = 70 : 30
Normal Phase	Halogen or acid–free organic solvent (hexane : ethanol = 90 : 10)
Ion Exchange Gel Permeation Hydrophobic Interaction	0.05% sodium azide solution and others
Column for Fullerene Separaton	Toluene and others
Sugar–D, NH_2, HILIC	Acetonitrile : water = 70 : 30

Attention;
Store tightly plugged columns in a vibration-free, cool dark place.

Q15. How long does a column last?

Column life time may change depending on the operation condition (sample, concentration, salt, acid, pH of organic solvent)
Attention;
The most common cause of a short column lifetime is inadequate sample treatment. Please refer to Technical Information 4 on page 193.

Q16. What happens when a column deteriorates?

Common symptoms of column deterioration include increasing column pressure, decreasing theoretical plates, shortening retention time, worsening of peak shape and decreasing resolution.

Attention;

Please refer to Troubleshooting on page 174.

Q17. How can I confirm the deterioration of column?

Evaluate deterioration of column under the same condition as the attached “certificate of analysis that comes with the column”. Use the same instrument every time to record the performance of the column. Record standard values over time and change to a new column if they are off.

Valued Item	Contents of Value
Capacity Factor (k ’)	If stationary phase is stripped, retention time may shorten.
Theoretical Plates (N)	It may decrease due to impurities or the state of packing material.
Peak Asymmetry (S)	It may decrease due to deterioration of packing material or adsorption of impurities.
Pressure	It may increase due to clogging of column filter, sample adsorption to packing material, compression of packing material and others.

(Reference value)

The following is reference value for 5C$_{18}$-MS-II (4.6 mm I.D. × 150 mm)

- Capacity factor (k ’) ：90% or less of Naphthalene’s k’
- Theoretical plates (N) ：9000 or less
- Peak asymmetry (S) ：out of range of 0.86–1.25
- Pressure ：20 MPa or more

Q18. What should I pay attention to when I use semi-micro columns?

Use injector, tubing and detector cell designed specifically for semi–micro column.

Attention;

Confirm linear mobile phase flow rate is in proportion to the column sectional area. For more information, please refer to Technical Information 2 on page 189.

Q19. What should I pay attention to when I use UHPLC columns?

- Use equipment for UHPLC.
- Shorten response of detector (e.g.,0.02 sec) when you use standard HPLC equipment.
- Use injector, tubing and detector cell designed for UHPLC columns.
- Column pressure should be 30 MPa or less.

Q20. How much sample can be loaded in a preparative column?

The sample load may differ depending on the sample resolution or solubility in mobile phase. Optimize maximum loading capacity by an analytical column, and determine maximum purified value in proportion to sectional area of column inner diameter.

Attention;

For scaling up from analytical column to preparative separation, please refer to page 35. For column inner diameter, flow rate and pipe, please refer to Technical Information 2 on page 189.

Q21. What should I pay attention to when I use both reversed phase and normal phase in the same instrument?

Replace solvent which both mobile phase can be mixed such as ethanol, then replace to new mobile phase.

Attention;

Mobile phase for reversed phase (e.g., methanol, water) and for normal phase (e.g., hexane) cannot be mixed.

Q22. What is the connection type of column?

Connection type of COSMOSIL and COSMOGEL is Waters type.

Attention;

Generally, waters type can be connected to most of instruments, but confirm the connection type with the manufacturer before using.

Q23. Which detection methods should I use?

Proper selection of a detector depends on the sample or the purpose of the experiment.

Detector	Feature	
Ultra Violet Visible Detector (UV/VIS detector)	[How to detect]	Detect sample absorbance.
	[Sample]	High sensitivity for compounds which have UV absorbance. Can not be used for compounds which do not have UV absorbance.
	[Feature]	Easy, widely used.
Fluorescence Detector (FLD)	[How to detect]	Detect fluorescence of photon excited sample.
	[Sample]	Fluorescent sample
	[Feature]	For sample which has little or no UV absorbance. High detection sensitivity enables trace component analysis.
Refractive Index Detector (RI detector)	[How to detect]	Detect difference in index of refraction between sample and mobile phase.
	[Sample]	All sample
	[Feature]	For sample which has little or no UV absorbance (e.g., saccharides, alcohols, amino acids). Its disadvantages include sensitive to change of temperature, component of mobile phase and flow rate.
Electro Chemical Detector (ECD)	[How to detect]	Detect electrochemically-active compounds such as oxidized or reducing compounds.
Evaporative Light Scattering Detector(ELSD)	[How to detect]	Detect scattering light of microparticulated target compound by evaporating mobile phase.
	[Sample]	Sample which has little or no UV absorbance (e.g., saccharides, alcohols, amino acids).
	[Feature]	For sample which has little or no UV absorbance (e.g., saccharides, alcohols, amino acids). It is not applicable for low-boiling compounds.
Mass Spectrometric Detector (MS Detector)	[Feature]	Able to measure MS spectrum of separated components for qualitative analyses. Disadvantage is compatible mobile phases are limited.

Q24. What is the common pressure unit?

Most of unit is in SI unit, MPa

Attention;

Old equipment sometimes have different unit (conversion：1 MPa = 10.197 kgf/cm^2= 145.0 psi = 10 bar).

Q25. What is dead volume?

Dead volume is the follow path volume from injector to detector not relevant to resolution.

Attention;

1. If dead volume is large, sample may spread and have poor peak shape.
2. Choose proper injector, detector cell and tubing , and column inner diameter to minimize dead volume.

For inner diameter of tubing or cell, refer to Technical Information 2 on page 189.

Q26. What is the difference between column pre-filter and guard column?

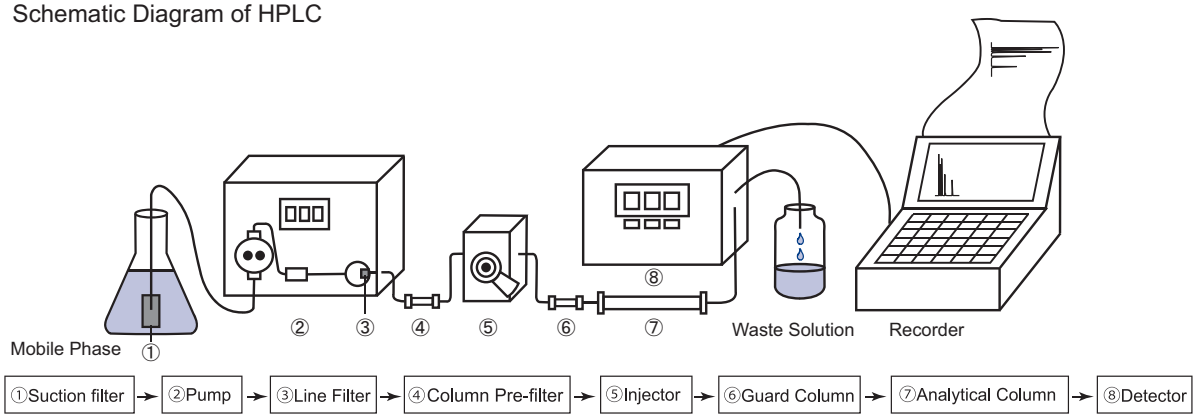
(1) Column pre-filter (④)

Connect between pump and injector to eliminate solid impurities in mobile phase.

(2) Guard column (⑥)

Connect between injector and analytical column to eliminate adsorptions in sample.

Schematic Diagram of HPLC



Q27. How can I pre-treat samples?

Refer to Technical information 4 on page 193.

Q28. How can I choose internal standards?

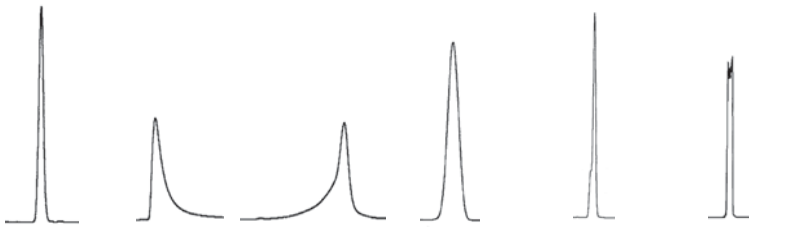
Ideal internal standards attribute,

- Peak of internal standards are near target compound peaks, but they do not overlap.
- Have no tailing or adsorption.
- Easy to procure at a low price.
- Have good chemical stability.

Methyl *p*-Hydroxybenzoate ~ Butyl *p*-Hydroxybenzoate is widely used in Japanese Phamacopoeia.

(3) Troubleshooting

T1. Poor peak shape



Normal Peak　Tailing Peak　Fronting Peak　Broad Peak　Peak Spike　Peak Splitting

Symptom	Cause	Solution
Particular sample is tailing	Undesirable ion exchange interaction between basic compounds and packing material.	Replace the column with less residual silanols (C_{18}-MS-II). Or add 0.1–1% of acid to mobile phase.
	Undesirable coordinate interaction between metal coordination compound and packing material.	Add 5mmol/l of di-sodium dihydrogen ethylenediaminetetraacetate dihydrate (EDTA・2Na) to mobile phase.
	Undesirable hydrogen bonding interaction between sample and packing material.	Change the organic solvent (e.g., acetonitrile to methanol).
All samples are tailing	Have spaces in packing material. Or column may have deteriorated.	Replace the column.
	(If the tailing does not improve after replacing the column) Sample is spreading out side of the column.	Reduce dead volume (Refer to Q25 on page 172 for more information on dead volume).
Fronting	Inject large volume of sample solvent that is significantly different in elution properties or pH comparing to mobile phase.	Dissolve sample in mobile phase. If the sample does not dissolve, dissolve in soluble solvent first, then dilute in mobile phase.
		Reduce injection volume to 1/2–1/10 Attention; Spikes or peak broadening may also occur.
Broad peaks 1 Sample has high molecular weight (MW: 2,000 or more).	Protein with high molecular weight cannot go into pores of packing material.	Use wide pore (pore size: 300A) column for reversed phase chromatography, COSMOSIL Protein-R. Refer to page 42 for more information.
	Sample volume is too large.	Reduce inject volume to 1/2–1/10. Attention; Tailing peaks may also occur.
	In case of broad tailing of a particular sample, compound may be adsorbed onto packing material.	Use COSMOSIL Protein-R, it has high recovery rate. Refer to page 42 for more information.
	In case of broad tailing of all samples, column may deteriorate.	Replace the column.
	Concentration of ammonium sulfate in sample solution is too low on hydrophobic chromatography (HIC).	Adjust concentration of ammonium sulfate to 1 mol/l or more.

Symptom	Cause	Solution
Broad peaks 2 Sample has low molecular weight (MW: 2,000 or less).	Sample volume is too large.	Reduce sample amount from 1/2 to 1/10. Caution; Tailing peaks may occur instead of broad peaks.
	In case of broad peak of a particular sample, compound may be adsorbed onto packing material.	Replace with a column that has a different packing material. (COSMOSIL 5C$_{18}$-MS-II is recommended for basic sample. The column has less adsorption to basic compounds. Please refer to page 14 for more information.)
	In case of broad peaks for all samples, the column may deteriorate.	Replace the column.
Peak spikes or peak splitting may occur for certain sample.	More than 2 samples are contained, and slightly separated.	Find the condition which enables separation of the two samples.
	Mobile phase and sample solvent are significantly different in their separation properties.	Dissolve sample in mobile phase. If sample does not dissolve, dissolve sample in sample soluble solvent first before mixing it with the mobile phase.
		Reduce loading capacity to 1/2 to 1/10.
	Mixed dissociated and non-dissociated ionic sample.	Adjust pH of mobile phase to pKa ± 2 or more of the ionic sample.
Peak spikes or peak splitting occur for all samples	Mobile phase and sample solvent are significantly different in their separation properties.	Dissolve sample in mobile phase. If sample does not dissolve, dissolve sample in sample soluble solvent first before mixing it with the mobile phase.
		Reduce loading capacity to 1/2 to 1/10.
	The column may have deteriorated.	Replace the column.

T2. Ghost peaks

Separation Mode	Cause	Solution
<Reversed Phase Chromatography> Use gradient elution method	Peaks from water impurities	Use new HPLC grade distilled water.
		Use a pre-column. Please refer to page 198 for more information.
<Reversed Phase Chromatography> Protein samples	Sample on previous analysis may be adsorbed onto the column, and elute on the next analysis.	Wash column, Please refer to page 170 for more information on washing methods.
		COSMOSIL Protein-R, which has high recovery rate for protein separations, is recommended. Please refer to page 42 for more information.
<All Separation Mode> Sample solvent and mobile phase are significantly different.	Sample solvent has peaks.	Dissolve samples in the same solvent as the mobile phase.
		Dissolve sample in mobile phase. If sample does not dissolve, dissolve sample in sample soluble solvent first before mixing it with the mobile phase.
<All Separation Mode> Mobile phase have peaks in blank analysis. (Peak area decreases with each injection.)	Injector is polluted	Wash column by injecting with a syringe of 20 ml solvent, e.g., methanol that can dissolve the pollutants
	Micro-syringe is polluted	Wash column with solvent e.g., methanol, chloroform or water to dissolve the pollutants. Ultrasonic cleaning is effective.
Others	Contamination or deterioration of samples	Adjust the sample again.
	Stabilizers in the mobile phase	Use HPLC grade solvent without stabilizers.

T3. No peaks

[How to confirm cause] Check t_0 first.

Analysis result of t_0	Cause
t_0 is not detected.	Detector may be defective.
Retention time of t_0 shifted.	Pump may be defective.
Retention time of t_0 is the same as usual.	Column may be defective.

● Solution for Each Column Type

Column Type	Cause	Solution
Reversed phase chromatography column	Sample is still in the column due to its high hydrophobicity.	Increase elution power of mobile phase until the sample elutes. e.g., 1. Increase concentration of methanol or acetonitrile (maximum 100%). 2. If the sample still does not elute, add 10–30% of higher elution organic solvent (e.g., tetrahydrofuran or chloroform) in methanol or acetonitrile (e.g., Tetrahydrofuran : methanol = 30 : 70).
	Metal coordination or basic compounds may be adsorbed onto the column.	Basic compounds may interact with residual silanols in the packing material. Use COSMOSIL 5C_{18}–MS–II, which has less residual silanols. Or add 0.1–1% of acid (e.g., trifluoroacetic acid, acetic acid) to the mobile phase.
		Metal coordination compounds may interact with a small amount of metal in the packing material. Add 5 mmol/l di–sodium dihydrogen ethylenediaminetetraacetate dihydrate (EDTA・2Na) to mobile phase.
Normal phase chromatography column	Hydrophilicity of sample is too strong so that sample is still inside of column.	Increase elution power of mobile phase until the sample elutes. Replace with strong eluting solvent e.g., ethanol, or increase the concentration
	Metal coordination or basic compounds may be adsorbed onto the column.	Add 0.1–1% of acid (e.g., trifluoroacetic acid, acetic acid) to mobile phase.
Column for saccharides analysis (Sugar–D, NH_2–MS) or Hydrophilic chromatography column (HILIC)	For COSMOSIL 5NH_2–MS, sample is adsorbed to amino groups.	Use COSMOSIL Sugar–D with less undesirable adsorption.
	Sample is still in column because it is higly hydrophilic.	Increase concentration of water in mobile phase until the sample elutes. COSMOSIL Sugar–D or COSMOSIL HILIC is compatible with 100% aqueous mobile phase. COSMOSIL 5NH_2–MS is compatible with 50% water (e.g., acetonitrile : water = 50 : 50).
Gel filtration chromatography column (Diol)	Sample has ionic effect on silanol group.	To increase ionic strength in mobile phase, add approx. 0.3 mol/l of salt, e.g., sodium chloride
		Adjust mobile phase pH to 5.5 or less to prevent ionic interaction.
	Sample may be adsorbed due to hydrophobic interaction.	Add 10–50% of organic solvent (e.g.,acetonitrile) to mobile phase.
Hydrophobic chromatography column (HIC)	Hydrophobicity of sample is too strong	Add 5% of organic solvent (e.g., methanol or acetonitrile).
	Sample may have ammonium sulfate precipitation before injection.	Decrease concentration of ammonium sulfate to 0.5mol/l or less until no precipitation is observed.

● Defective Pump

Cause	Solution
Bubbles are generated in a pump.	Collapse bubbles (Please refer to T7 on page 180.).
Solvent leaking	Tighten connectors or replace tubings.

● Defective Detector

Cause	Solution
Detector is not connected correctly.	Follow the detector user's manual to connect correctly.
Defective signal from the detector	Contact detector manufacturer
UV adsorption range is not suitable for your sample.	Analyze at suitable UV adsorption for a sample. If the sample has no or little UV adsorption, use refractive index detector (RI detector) or evaporative light scattering detector (ELSD), or labeling the sample.

T4. Unstable base line

Cause	Solution
Impurities adsorbed onto a column previously are eluting out now.	Wash with strong eluting solvents (Please refer to Q13 on page 170.).
COSMOSIL PE-MS • π NAP • PYE • NPE • PBB-R • Cholester have UV absorption on stationary phase, and base line is not stable due to slight shedding (the detachment of stationary phase that has UV absorption.).	Wash with strong eluting solvents (Please refer to Q13 on page 170.).
Sudden change in the pump pressure may create bubbles.	Degas (Please refer to T7 on page 180.).
When using refractive index detector (RI detector)	
large temperature variations	Use thermostatic bath to keep a constant temperature. Beware of the air conditioner blowing on the RI detector or tubing. Caution; Cover equipment or tubing to avoid temperature fluctuation from an air conditioner.
Residual gas in mobile phase changes.	Degas (by ultrasonic wave or aspirator) a mobile phase.
Have needle-like peaks from an ultra violet visible detector	
Bubbles may be mixed in the column or detector.	Increase pressure to remove bubbles by blocking exit of the detector. Caution; Too much pressure may break the detector cell. If the problem persists, run thick solvent (e.g., 2-propanol) through for 15 min, disconnect the column from the detector.
Column temperature may be above the boiling point of mobile phase, creating bubbles in a column.	Analyze at suitable temperature. Basically, 20-50°C is suitable temperature for a column. Caution; To get the best result, analyze at 20-50°C lower than the boiling point of mobile phase (e.g., in case of Methanol [boiling point: 64.7°C], analyze at 45°C or less.).
When using ion-pair reagent for mobile phase or buffer.	
Inadequate equilibration of a column	Make equilibration time longer. When using ion-pair reagent for mobile phase or buffer, longer equilibration time is required compare to mobile phases without salt.
Salt may precipitate in the mobile phase and the mobile phase reservoir may become cloudy.	(a) Decrease concentration of buffer. (e.g., 100 mmol/l → 20 mmol/l) (b) Replace with a different buffer solution. (e.g., phosphoric acid buffer → acetic acid buffer) (c) Reduce concentration of organic solvent. (e.g., 70% acetonitrile : water = 70 : 30 → 50 : 50) (d) Replace with a different organic solvent. (e.g., acetonitrile → methanol)

T5. Unstable retention time

● Cause by Equipments

Equipment	Cause	Solution
Pump	Bubbles in the check valve of pump.	Degas (Please refer to T7 on page 180.).Normal phase solvents have lower boiling point, so bobbles are easily created. Furthermore, its low-viscosity prevents bubbles from eluting.
	Solvent leaking	Tighten the leaking part. If the problem does not solve, replace it.
Thermostatic Bath (for adjusting temperature)	Column temperature may vary by season or the time of day without the use of thermostatic bath or column oven.	Use thermostatic bath or column oven to keep consistent column temperature. Caution; Set thermostatic bath or column oven to 5°C above the room temperature when doing room-temperature analyses.

● Cause by Columns

Column Type	Cause	Solution
Reversed Phase Column	Inadequate column equilibration when using ion paring reagents.	Make equilibration time longer. When using ion-pair reagents, longer equilibration time is often required.
	If 100% water is used as mobile phase on C_{18} column, phase collapse may occur.	COSMOSIL C_{18}-PAQ is compatible with 100% aqueous mobile phase (Please refer to page 18.). Caution; For unstable retention time, wash the column with high organic solvents (e.g., methanol : water = 70 : 30) to recover.
Normal Phase Column	A small amount of water in organic solvent may affect retention time.	Replace with mobile phase without water. If sample solvent has water, change sample solvent or decrease the injection volume. If water is trapped, wash with ethanol to recover.
Columns for Saccharide Analysis (Sugar-D, NH_2-MS) or Hydrophilic Chromatography column(HILIC)	A small amount of stationary phasedetached.	COSMOSIL Sugar-D or COSMOSIL HILIC can be recovered by washing with 100% water for 15 minutes. COSMOSIL 5NH_2-MS may be recovered by washing with 50%water (e.g., acetonitrile : water = 50 : 50) for 15 minutes.

T6. Increased column pressure

Please refer to Technical Information 3 on page 191.

T7. Unstable pump pressure

Cause	Solution
Bubbles in the check valve of a pump.	Degas the check valve (open drain valve, and let mobile phase through) according to the pump instruction. If the problem persists, wash check valve, e.g., ultrasonic cleaning in water.

Caution;

1. If the bubbles occur often in normal phase chromatography, connect pre-column to increase pressure, and let bubbles elute.
2. Degas a mobile phase by a ultrasonic or an aspirator.

T8. Poor resolution on C_{18} columns

Solution	Features
Use a longer column	A longer column enables sharper peaks. The pressure will increase and the retention time will be longer.
Change mobile phase condition (e.g., pH, type or concentration of organic solvent)	Experience or knowledge is required. Do not set complicated condition as it lacks repeatability.
Use packing material with non-hydrophobic interactions.	Separate sample by molecular shape selectivity or π-πinteraction (except for hydrophobic interaction). Please refer to Technical Information 7 on page 201 for more information on special column with various interactions.

T9. No retention on reversed phase columns

Solution	Features
Use ion pair reagents	Ion pair reagents enable separation by forming ion pairs with the sample to increase hydrophobicity. Therefore, it is not applicable for non-dissociative samples.
Use hydrophilic columns (HILIC). Please refer to page 36.	Less hydrophobic samples are retained longer.

T10. Excessive retention time is long on reversed phase columns

Solution	Features
Use the gradient elusion method	Gradient elution method shortens analysis time by changing organic solvent concentration during analysis. Disadvantages are having a capable equipment, increasing baseline, and the need for equilibration time between each runs.
Use UHPLC Columns	Please refer to page 62, for more information.
Change mobile phase condition	Problem may be solved by changing pH, type or concentration of organic solvent.
Use column with small hydrophobicity	COSMOSIL CN-MS is recommended. Please refer to page 31, for more information.

T11. Different separation performance compare to the past

Symptom	Cause	Solution
Decreased theoretical plates	Natural deterioration of the packing material	No method to recover column.
Decrease of retention time or separation.	Impurities may be adsorbed onto the packing material.	The column can be recovered by washing
	Stationary phase shedding.	No method to recover column.

T12. Different separation performance with a new column

Cause	Solution
Analytical condition does not suit to sample	Adjust pH of mobile phase to pKa ±2 or more.
	Use mobile phase with high repeatability.
Column may deteriorate	If the column deteriorate, decrease of retention time, change of peak shape may occur. Replace the column.
Variation from lot to lot	Contact us with the column name, product no, present separation status. (a) Evaluate columns with 3 different packing material lots. We provide validated column with 3 different packing material lots for the following columns (Size 4.6 x 150 mm, 3 pkg set). Contact us for more information. Packing material / Product No. COSMOSIL 5C_{18}-MS-II / 09397-73 COSMOSIL 5C_{18}-AR-II / 09396-83 COSMOSIL Cholester / 07970-03 COSMOSIL HILIC / 09385-23 (b) Find an analysis condition with less influence from lot-to-lot variation.
Cause is not column (e.g., mobile phase, flow rate, temperature)	Find the cause.

T13. Colored elute from columns (colorless sample)

Cause	Solution
A small amount of shedding, impurities or previous samples.	Wash column with strong elution solvent (e.g., methanol) or use cleaning solution kit for reversed phase HPLC columns (Product No. 08966-30)

Caution;
A small amount of stationary phase does not affect retention time.

T14. Air got inside the column (dried out)

Pump solvent with low viscosity (e.g., methanol) through at half of analysis flow rate for 1 hour.

Caution;

Store it tightly plugged in a cool dark place.

2. Liquid Chromatography Basics

History of Liquid Chromatography

Liquid chromatography was defined by Mikhail S. Tswett (1906–1907) who separated leaf pigments into different colored bands using chalk powder ($CaCO_3$) as adsorbent. Then reversed phase chromatography, ion exchange chromatography, size exclusion chromatography were developed. In 1971 J.J. Kirkland has succeeded in the production of chemically bonded packing material for liquid chromatography, and contributed to establish the basics of high-performance liquid chromatography, which are now one of the most important analysis methods.

HPLC Equipments

HPLC equipments are connected as shown in Figure 1 in the order of mobile phase flow from mobile phase reservoir, pump, injector, column, detector to waste solvent container. Samples are introduced into mobile phase through the injector and separated by the column. Chromatogram is drawn by recorder.

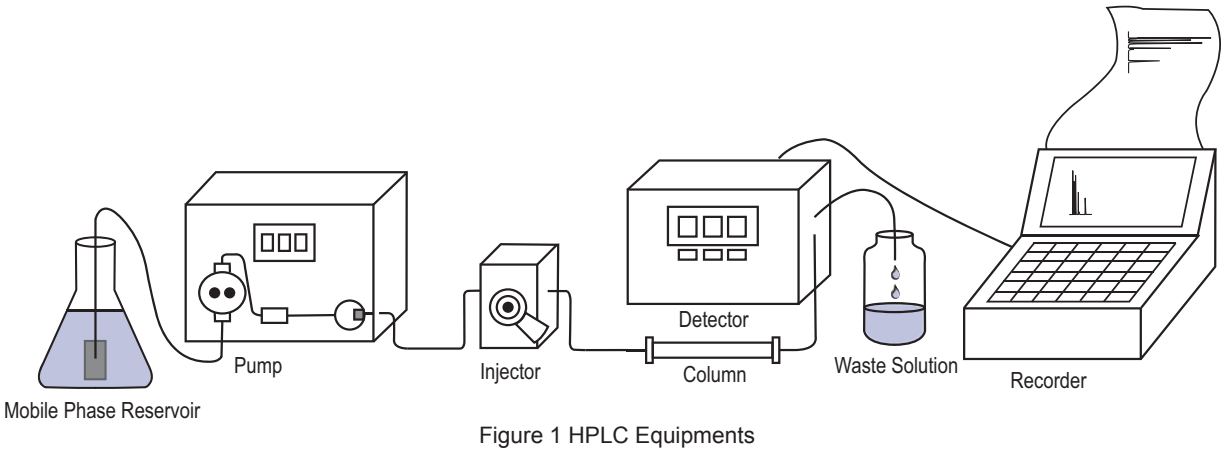


Figure 1 HPLC Equipments

- Mobile Phase Reservoir
  Glass bottle or conical flask is used as a mobile phase reservoir. To avoid clogging from insoluble compounds in the mobile phase, a suction filter (or sinker) is attached to the inlet.
- Pump
  Send mobile phase at a consistent flow rate or pressure. Connecting two pumps enables gradient elution.
- Injector
  Inject sample to column by a micro-syringe. Auto-injector is widely used for automated sample injection.
- Column
  Packing material is packed in a stainless or glass chromatogram column. To avoid the elution of packing material, a frit (2 μm) is packed on each end of the column.
- Column Thermostatic Oven
  Maintain the column at a consistent temperature. Temperature control is very important for reversed phase chromatography and ion exchange chromatography. It is desirable to keep the temperature within ±0.5°C. Water or air circulator is widely used.
- Detector
  Detect each compounds eluted from the column, and convert them into electronic signals.
- Recorder
  Process the electronic signals from detector to draw chromatograms. Retention time, peak area, and theoretical plate numbers are automatically calculated.

Chromatogram

Elution means the process to extract retained samples from a column. Retention time means the time between the sample injection and the sample extraction. Chromatogram is a two-dimension diagram (Figure 2) that reveals the retention time on the abscissa and the concentration of solute on the ordinate. The chromatogram shows following data.

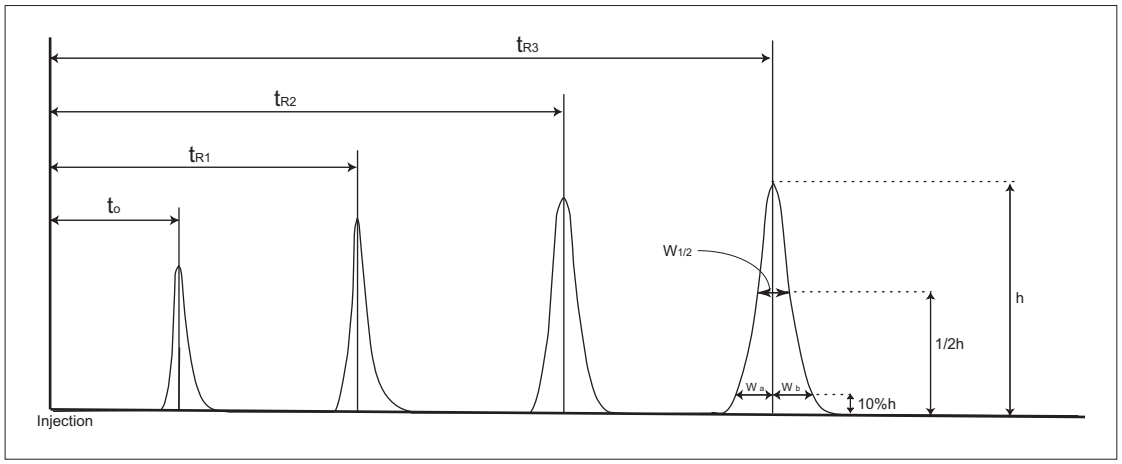


Figure 2 Chromatogram

(1) t_o : Retention time of the mobile phase

The retention time of an unretained peak. Uracil is widely used in reversed phase as a t_o marker.

(2) t_R : Retention time

Distance from the point at peak midpoint to the start of analysis.

(3) h : Peak height

Distance from the peak top perpendicular to the baseline.

(4) $W_{1/2}$: Peak half width

The peak width at half height

(5) k' (capacity factor) : Capacity factor, retention ratio of each sample, $k' = (t_R - t_o)/t_o$

A higher volume means longer retention. . The value remains consistent under the same experimental condition (packing material, mobile phase and temperature)

(6) N (theoretical plate) : theoretical plate number, $N = 5.54 (t_R/W_{1/2})^2$

A theoretical plate is an imaginary layer within a column that helps to interpret the separation process. A higher theoretical plate number corresponds to better column efficacy. The plate number depends on the packing material and experimental conditions.

(7) S (peak asymmetry) : peak asymmetrys, $S = W_b/W_a$

Wa : Distance from the leading edge of the peak to the midpoint (measured at 10% of peak height)
Wb : Distance from the point at peak midpoint to the trailing edge (measured at 10% of peak height)
The peak asymmetry S=1 indicates a perfectly symmetrical peak, and S > 1 indicates tailing, and S < 1 indicates leading. Tailing or leading occurs with deteriorated packing material, unsuitable experimental conditions or overloading.

(8) α (separation factor) : Separation factor, $\alpha = k'_2/k'_1$ (k'_1 and k'_2 : retention ratio of each sample)

The separation factor must be > 1 for peak separation. A higher α value indicates greater distance between the peaks.

(9) Rs (resolution) : Resolution, $Rs = \{\sqrt{N}/4\} \cdot (\{\alpha-1\}/\alpha) \cdot \{k'_2/(1+k'_2)\}$

The resolution (Rs) indicates how well two samples are separated. Rs=1.5 indicates baseline separation. If the Rs value is smaller than 1.5, peaks may overlap.

Features of Each Separation Mode of HPLC

HPLC has following separation modes.

● Normal Phase Chromatography

Adsorbent material such as silica gel or alumina is used as the packing material. Analytes are separated by the difference in adsorptive forces to the packing material, resulting in each moving at different speed. The analyte that interacts more strongly with the packing material moves at a slower rate.

● Reversed Phase Chromatography

Analytes are distributed between polar mobile phase and non-polar stationary phase, and separated by the flow speed difference due to the difference in the distribution between these two phases. If the analyte distributes more to the stationary phase, it would have a slower flow speed. Non-polar packing materials, such as octadecyl group and octyl group bonded silica gels are widely used. They are stable to heat and hydrolysis within a certain pH and temperature range, separation is influenced by the type of stationary phase, carbon rate, end capping treatment, etc.

● Ion-exchange Chromatography

Charged functional groups are bonded to the solid support to separate ionic solutes with the counter-ions. Analytes are separated by the flow speed difference due to the difference in the affinity to stationary phase. Dextran, cellulose and polystyrene are commonly used as the packing materials. Typical functional groups are Sulfopropyl (SP) and Carboxymethyl (CM) for cationic exchange, and Diethylaminoethyl (DEAE) and Quarternary ammonium (QA) for anionic exchange. The ion exchange capacity which influences separation performance depends on the type and density of the functional groups.

● Size Exclusion Chromatography

Analytes are separated by the molecular size. The analyte smaller than the pore size can penetrate the pores and migrate slowly, whereas larger analyte is excluded from the pores and migrates quickly. This mode mainly used for separation of high molecular polymer (molecular weight 2,000 and more). Organic expanded type gels (such as dextran and polyacrylamide) and inorganic gels (such as silica gel and glass) are used as packing materials.

Separation Mechanism of HPLC

The most common mode, reversed phase is used as an example here. Mixed samples are injected into a column, the lower hydrophobic analyte (A) distributes in the polar mobile phase and move faster down the column. Conversely, higher hydrophobic analyte (B) distributes in the non-polar stationary phase for a longer time and moves slower down the column. Therefore, the analytes flow out of the column in order of the polar first and the non-polar last.

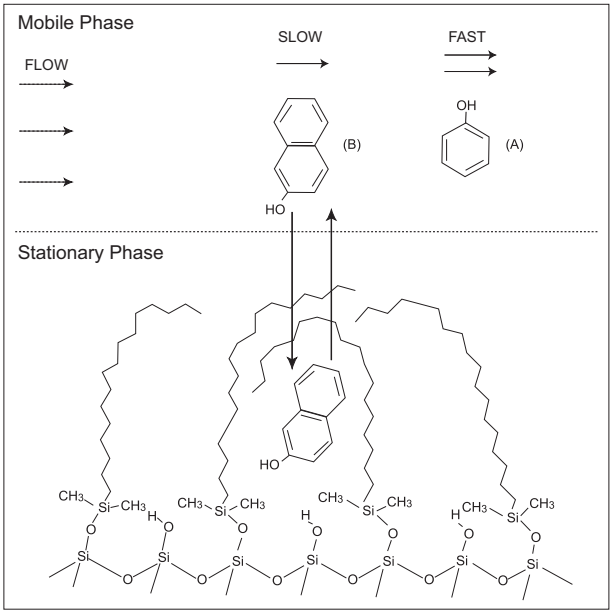


Figure 3. Separation Mechanism

Mobile Phase Solvent

The important mobile phase qualities for HPLC are shown below.

1. High solubility for the sample components
2. Good miscibility
3. No detection disturbance
4. Low viscosity
5. Higher boiling point than the operational temperature
6. Low toxicity and non-flammability
7. Low price
8. Using HPLC grade or filtered solvents.

Next, selection guide of mobile phase is shown below according to the separation mode.

● Normal Phase Chromatography

Generally, a polar solvent is mixed with a nonpolar solvent. The separation factor is adjusted by changing the mixed ratio. Please refer to the polarity and the solubility of solvent. Toluene, hexane, chloroform, ethyl acetate, and ethanol are mainly used.

● Reversed Phase Chromatography

Water, methanol, acetonitrile, and tetrahydrofuran are mainly used. Separation factor is adjusted by the mixed ratios of these solvents. When using silica-based columns for ionic analytes, it is desirable to adjust the pH range from 2 to 7.5. Generally, silica-based columns are not stable outside of this range due to cleavage of the bonded groups at pH < 2, and the dissolution of silica support at > pH 7.5. Use filtered phosphoric acid buffer solution or acetic acid buffer solution for pH control.

● Ion-exchange Chromatography

Add buffer solution in water and adjust separation factor by salt concentration (ionic strength) and pH. The more ionic strength is, the earlier the sample elutes. Lower pH decreases the separation factor on anion exchange, and increases the one on cation exchange. Cation buffer solutions, such as ammonia and amine are used for anion exchange, and anion buffer solutions, such as acetic acid salt, formic acid salt and citric acid are used for cation exchange.

● Size Exclusion Chromatography

Generally, a single solvent is used as the mobile phase, and it is not changed to adjust the separation factor. Tetrahydrofuran, chloroform, toluene anddimethylformamide are commonly used in non-aqueous mode. Add buffer solution in water for aqueous mode. Adjust pH and ionic strength to prevent adsorptionand other undesired interactions.

Quantitative Analysis

Absolute calibration curve method or internal standard method is used to calculate the amount or concentration of solute by peak area or height.

● Absolute Calibration Curve Method

1. Prepare the standard solutions in 3–4 different concentrations.
2. Inject the same volume of each standard solution, record chromatogram, and measure peak area.
3. Prepare a calibration curve by plotting the amounts of the standard on the x–axis and the peak areas on the y–axis. The calibration curve is usually a straight line through the origin.
4. Inject sample under the same conditions as the standards, and record a chromatogram. Measure the peak area (y) and use the calibration curve to determine the sample amounts.

This method should be performed exactly under a given condition. This method is also called external standard method.

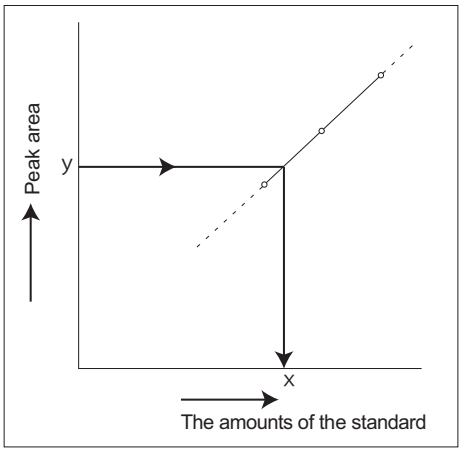


Figure 4. Absolute calibration curve

● Internal Standard Method

1. Prepare 3–4 known concentration*1 ratios of the standards and samples*2.
2. Inject a constant volume of each concentration, record chromatogram and measure peak areas.
3. Prepare a calibration curve (as shown on fig. 5) by plotting M_X/M_S vs. A_X/A_S ratios. M_X is the amount of the sample injected, and M_S is the amount of the standard. A_X is the peak area of the sample, and A_S is the peak area of the standard. The calibration curve is usually a straight line through the origin.
4. Then, prepare a the test solution containing a known amount of the internal standardand an unknown amount of sample*3. Perform the experiment under the same conditions as for obtaining the calibration curve.
5. Use the calibration curve to determine the unknown sample amount.

*1 If the calibration curve is confirmed to be a straight line through the origin, plot the calibration curve with AX/AS determined by one point of concentration of injected unknown sample.

*2 The internal standard should have similar chemical characters as the sample while completely separated from it.

*3 When the internal standard is added to the test solution, make sure the chemical reaction (e.g., precipitation) does not occur.

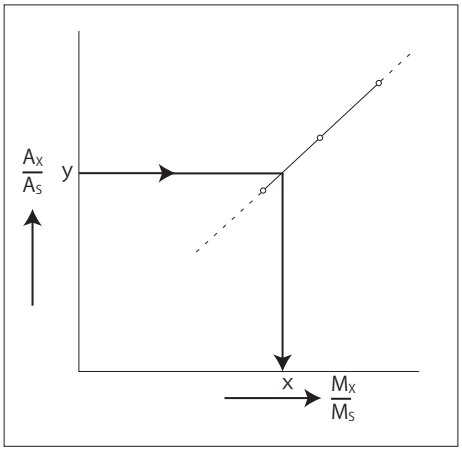


Figure 5. Calibration Curve of Internal standard method

1. Preparation of Mobile Phase for HPLC

1) Organic Solvent / Aqueous Mixed Mobile Phase

(e.g.) Methanol : Water = 70 : 30 1L

Prepare mobile phase by volume ratio.

1. Measure 700 ml of methanol in a measuring cylinder.
2. Measure 300 ml of distilled water in a measuring cylinder.
3. Mix 1 and 2 thoroughly and degas.

Attention; The better approach is to prepare the mobile phase gravimetrically rather than volumetrically. Following is example of preparation.

Composition table for mobile phase 1L (Methanol : water)

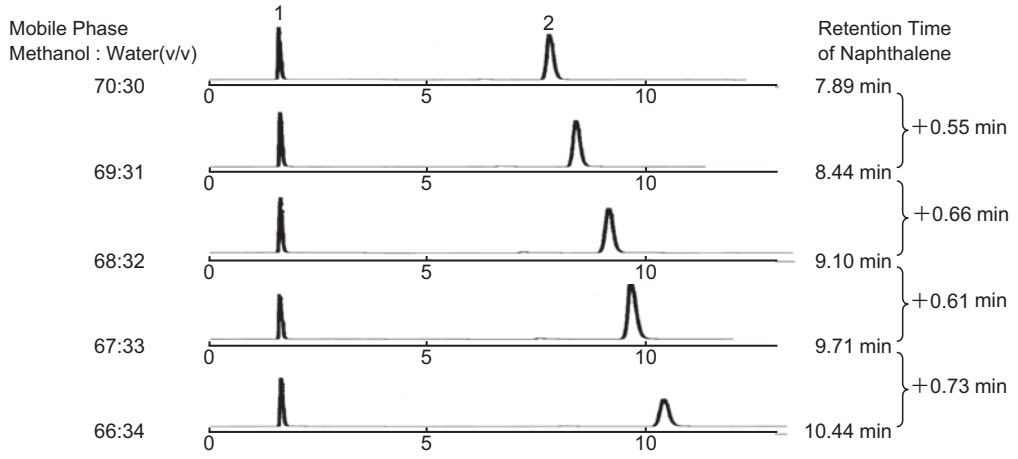
Methanol / Water	Methanol (g)	Distilled Water (g)
90 : 10 (v/v)	711.9	99.8
80 : 20 (v/v)	632.8	199.6
70 : 30 (v/v)	553.7	299.5
60 : 40 (v/v)	474.6	399.3
50 : 50 (v/v)	395.5	499.1
40 : 60 (v/v)	316.4	598.9
30 : 70 (v/v)	237.3	698.7
20 : 80 (v/v)	158.2	798.6
10 : 90 (v/v)	79.1	898.4

Composition table for mobile phase 1L (Acetonitrile : water)

Acetonitrile / Water	Acetonitrile (g)	Distilled Water (g)
90 : 10 (v/v)	707.4	99.8
80 : 20 (v/v)	628.8	199.6
70 : 30 (v/v)	550.2	299.5
60 : 40 (v/v)	471.6	399.3
50 : 50 (v/v)	393.0	499.1
40 : 60 (v/v)	314.4	598.9
30 : 70 (v/v)	235.8	698.7
20 : 80 (v/v)	157.2	798.6
10 : 90 (v/v)	78.6	898.4

Caution : Methanol and acetonitrile are hazardous substances, do not use for medical purpose. Always process in a laboratory hood and wear an eye protection and a mask.

(Reference) Influence of organic solvent composition in mobile phase on the retention time.



Special attention should be paid to measure correct amount of organic solvent as retention time is significantly changed by 1% different composition.

Column	5C18-MS-II 4.6 mmI.D. x 150 mm
Flow rate	1.0ml/min
Detection	254nm 0.16AUFS
Temperature	30°C
Sample	1. Uracil 2. Naphthalene

2) Organic Solvent / Buffer Mixed Mobile Phase

(e.g.1) Preparation of 20 mmol/l phosphate buffer (pH2.5)

1. Preparation of 20 mmol/l sodium dihydrogenphosphate aqueous solution (Dissolve 2.40g of sodium dihydrogenphosphate, Anhydrous (Product No. 31720-65) in distilled water to make 1L solution.)
2. Prepare 20 mmol/l phosphate aqueous solution (Dissolve 2.31g of Phosphoric acid (Purity: 85%), (Product No. 08964-92) in distilled water to make 1L solution.).
3. Adjust the pH to 2.5 by mixing 1 with 2.
4. Filter under reduced pressure to remove insoluble substance (0.45 µm or smaller pore size is recommended.). Attention; Filter solids from the solution to prevent clogging to pump and columns.
5. When mix with organic solvent, mix by volume ratio.
   Attention; The solid may precipitate after mixing.

For more information on adjusted solution, Phosphate Buffer Solution (pH 2.5) (5x) (Product No, 08969-71), Please refer to page 77.

(e.g.2) Preparation of 20 mmol/l phosphate buffer (pH7.0)

1. Preparation of 20 mmol/l sodium dihydrogenphosphate aqueous solution (Dissolve 2.40 g of sodium dihydrogenphosphate, Anhydrous (Product No. 31720-65) in distilled water to make 1L solution.)
2. Prepare 20 mmol/l di-sodium hydrogenphosphate aqueous solution (Dissolve 2.84 g of di-Sodium Hydrogenphosphate, (Product No. 31801-05) in distilled water to make 1L solution.).
3. Adjust the pH to 7 by mixing 1 with 2.
4. Filter under reduced pressure to remove insoluble substance (0.45 µm or smaller pore size is recommended.). Attention; Filter solids from the solution to prevent clogging to pump and columns.
5. When mix with organic solvent, mix by volume ratio.
   Attention; The solid may precipitate after mixing.

For more information on adjusted solution, Phosphate Buffer Solution (pH 7.0) (5x) (Product No, 08968-81), Please refer to page 77.

(e.g.3) Preparation of 5 mmol/l Sodium *1*-hexanesulfonate, 20 mmol/l phosphate buffer (pH2.5)

1. Prepare 5 mmol/l Sodium *1*-hexanesulfonate, 20 mmol/l phosphate buffer (pH2.5) aqueous solution (Dissolve 10 ml of Sodium *1*-hexanesulfonate (0.5 M solution) (Product No. 31532-06) and 2.40 g of sodium dihydrogenphosphate, Anhydrous (Product No. 31720-65) in distilled water to make 1L solution.).
2. Prepare 5 mmol/l Sodium *1*-hexanesulfonate, 20 mmol/l phosphate aqueous solution (Dissolve 10 ml of sodium *1*-hexanesulfonate (0.5 M solution) (Product No. 31532-06) 2.31g of phosphoric acid (Purity: 85%), (Product No. 08964-92) in distilled water to make 1L solution.).
3. Adjust the pH to 2.5 by mixing 1 with 2.
4. Filter under reduced pressure to remove insoluble substance (0.45 µm or smaller pore size is recommended.). Attention; Filter solids from the solution to prevent clogging to pump and columns.
5. When mix with organic solvent, mix by volume ratio.
   Attention; The solid may precipitate after mixing.

2. Inner Diameter of Column (scale down and scale up)

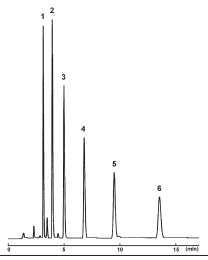
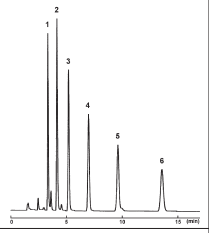
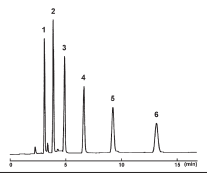
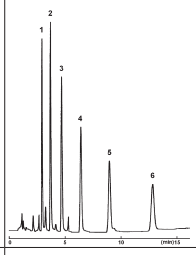
Introduction

The figure below shows general parameters for 1.0 mm to 50 mm I.D. COSMOSIL columns : flow rate, equipment, inner diameter of pipe, application, surface ratio (compared with 4.6 mm I.D.) and particle size. It may help to scale up or down from the most commonly used 4.6 mm I.D. column.

Inner Diameter (mm I.D.)	1.0	2.0	3.0	4.6	10	20	28	50
Flow Rate (ml/min)	0.05	0.2	0.4	1.0	5.0	19	37	70
Detector Cell・Injector	for Semi-micro		for Analytical			for Preparative		
Inner Diameter of Pipe (mm)	0.05	0.1	0.2-0.3			1.0		
Application	LC-MS Solvent saving		Solvent saving with standard system	Standard	Preparative (small scale)	Preparative (medium scale)	Preparative (large scale)	Preparative (super large scale)
Surface Ratio with 4.6 mm I.D.	0.05	0.19	0.43	1.00	4.73	18.90	37.05	118.15
Particle Size (μm)	3 or 5				5		15 or more	

Scale Down

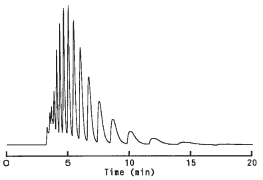
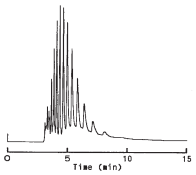
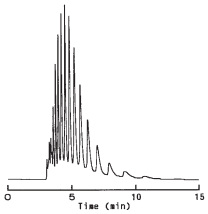
When scaling down from the most commonly used analytical column (4.6 mm I.D.) to a semi-micro or 3.0 mm I.D. analytical HPLC column (of the same column length), sample loading dose is proportionate to the cross section of column. The 3.0 mm I.D. columns provide high sensitivity and solvent saving without the need to change the existing equipment settings. Semi-micro columns (2.0 mm I.D. and 1.0 mm I.D.) provide higher sensitivity and enable analysis of minor components, but one needs to change the piping of HPLC equipment, the injector and the detector cell for semimicro columns.

Column Size	4.6 mm I.D. x 150 mm	3.0 mm I.D. x 150 mm	2.0 mm I.D. x 150 mm	1.0 mm I.D. x 150 mm
Chromatogram				
Flow Rate (ml/min)	1.0	0.4	0.2	0.05
Pressure (MPa)	3.4	3.6	3.8	3.6
Injection Volume (μl)	1.0	0.4	0.2	0.05
Detector Cell・Injector	for Analytical		for Semi-micro	
Detector sensitivity (AUFS)	0.08		0.04	
Inner diameter of pipe (mm)	0.25		0.10	0.05

Column	COSMOSIL $5C_{18}$-MS- II	Sample	
Mobile Phase	Acetonitrile : Water = 70 : 30	1. Benzene	4. Propylbenzene
Flow Rate	1.0 ml/min	2. Toluene	5. Butylbenzene
Temperature	30°C	3. Ethylbenzene	6. Amylbenzene
Detection	UV 254 nm		

Scale Up

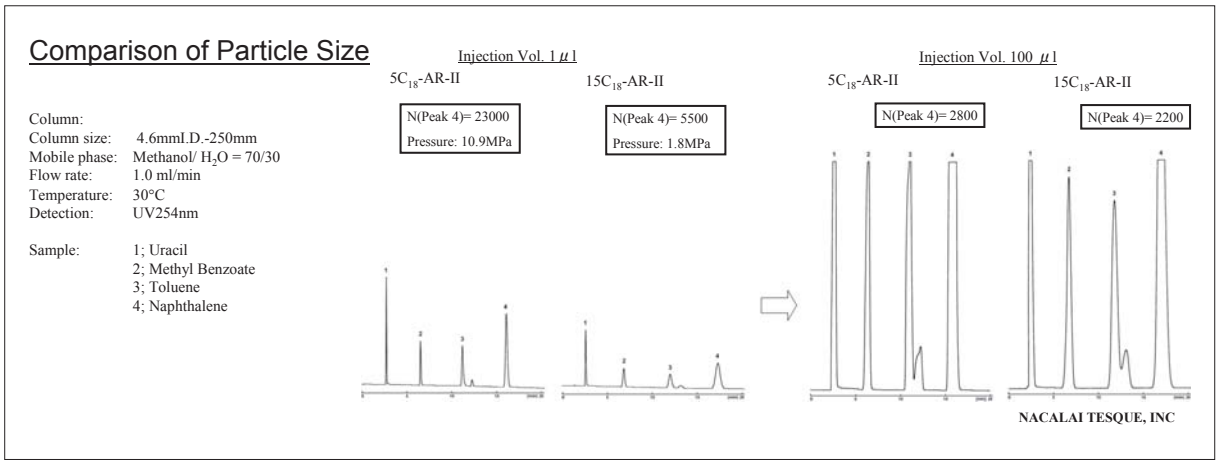
When scaling up from analytical column (4.6 mm I.D.) to preparative column (of the same packing material (particle size) and length), sample loading capacity is proportionate to the cross section of column.

Column Size	4.6 mm I.D. x 250 mm	10 mm I.D. x 250 mm	20 mm I.D. x 250 mm
Chromatogram			
Flow Rate (ml/min)	1.0	5.0	18.9
Pressure (MPa)	5.5	5.9	5.8
Injection Volume (µl)	125	625	2,500
Detector Cell・Injector	for Analytical		Preparative
Inner Diameter of Pipe (mm)	0.25		1.0

Column COSMOSIL 5SL-II
Mobile Phase Ethyl Acetate : Ethanol = 4 : 1
Temperature 30°C
Detection UV 254 nm
Sample Triton X-100

Comparison of Particle Size

When change particle size of packing material from 5 µm to 15 µm, the number of theoretical plate (N) is reduced by one-third, and the pressure is reduced by one-ninth. As shown in the figure below, when a small amount of sample is injected, there is a big difference in the number of theoretical plates between 5 µm and 15 µm. However, when a large amount of sample is injected, there is not much difference between the two. Therefore, the low pressure packing material (particle size 15 µm) is recommended for preparative column (28 mm I.D. or more).



3. Troubleshooting for Increased Pressure

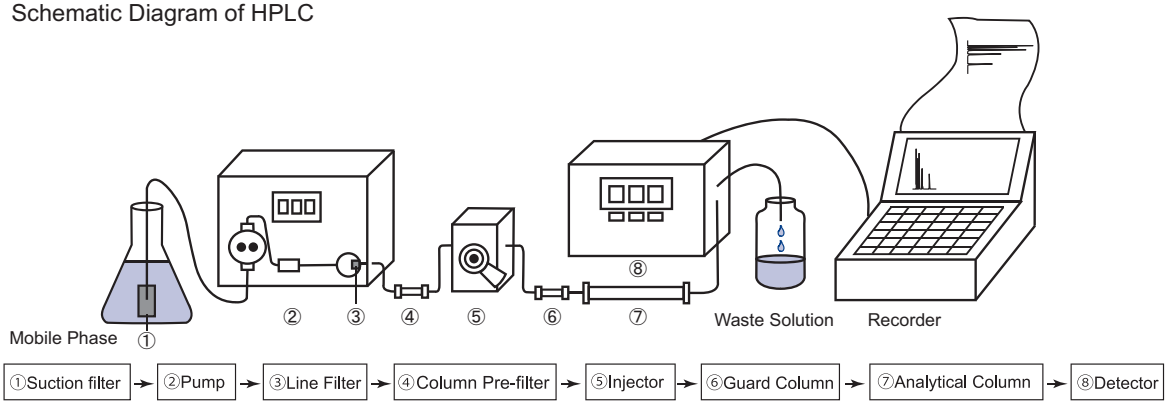
Introduction

Repeated analysis may increase back pressure. Continuous use of HPLC columns under high pressure can cause deterioration and overload of the equipment. Therefore, it is important to monitor column back pressure regularly and solve the problem timely.

Identification of the Clogging Site

The back pressure increase can be due to clogging of a column or clogging of the equipment. First of all, identify the clogging site.

Schematic Diagram of HPLC



Disconnect the system components one by one to identify the clogged component(s). Start by disconnecting the column from the system and measure the pressure of flowing mobile phase. The pressure should be close to zero. If the flow pressure without a column is normal, then pressure increase is due to the clogged column. The cause of clogging needs to be determined and preventative measures implemented. The column may need to be replaced.

Clogging of Equipment

Identify the specific clogging site according to the method above.

(Case 1) High pressure caused by clogged tubing

Cause : Salt deposit in tubing.

Solution : Disconnect the column and any other equipment before pumping water through the tubing. Washing in a reversing direction is also an effective way. If the situation does not improve, replace the tubing with a new one.

(Case 2) High pressure caused by clogged pump

Cause : Line filter of pump is clogged.

Solution : Take apart the line filter, and soak it in the solvent, then clean in an ultrasonic cleaner. If the situation does not improve, replace the line filter with a new one.

(Case 3) High pressure caused by clogged manual injector

Cause : Manual injector is clogged.

Solution : Inject with 20ml of contaminant dissolving solvent (e.g., methanol) by syringe. Wash both lines in LOAD and INJECT position. Cleaning the injector in an ultrasonic bath is also effective. If solids caused the clogging, wash the injector in a reversing direction. If the situation does not improve, replace the injector with a new one.

What should I do when a clogged column caused pressure increase?

(Case 1) Salt deposit in a column caused by pumping high-organic solvent after using buffer solution

Cause : Salt deposit in a column.

Solution : Wash columns for 30 minutes at half the normal flow rate using 10% organic solvent (methanol or acetonitrile) in water to dissolve salt deposit. If the situation does not improve, wash the column with 100% water under the same condition.

Prevention : To switch to high organic solvent concentration after using a buffer, first wash a column with a salt-free mobile phase (with the same concentration of organic solvent as the buffer), then switch to the mobile phase of higher organic concentration.

Example : To change mobile phase from 10/90 (v/v) acetonitrile/20mmol/l phosphate buffer (pH2.5) to 90/10 (v/v) acetonitrile/water, first wash the column for 15 minutes with 10/90 (v/v) acetonitrile/water, and then switch to 90/10 (v/v) acetonitrile/water.

(Case 2) The sample is not completely dissolved or unfiltered

Cause : Column frit is clogged by insoluble sample or impurities.

Solution : Connect the column in the reverse direction and disconnect from the detector, and then wash the column for 30 minutes at half of the usual flow rate with the same mobile phase used for analysis. If the situation does not improve, change the frit in the front end of the column (We can replace end fittings with a paid service fee.).

Prevention : We strongly recommend filtering sample and/or mobile phase. For more information, please see page 193, Technical Information 4. Sample Pretreatment for HPLC 1) filtration.

Attention : If the column is continually connecting in the reverse direction, it may deteriorate.

(Case 3) Protein samples that adsorb easily to the column or samples that are slightly soluble in mobile phase

Cause : Samples have adsorbed to packing material or deposited in a column.

Solution : Wash the column for 30 minutes with half of the normal flow rate using a solvent that can dissolve the adsorbed substances. Here are washing procedures for each column type.

[Reversed phase columns]

a) When absorbed substances are not proteins, wash with methanol or tetrahydrofuran.

b) When absorbed substances are proteins, wash with 50-70% of acetonitrile/water (containing 0.1% of trifluoroacetic acid). However, proteins may precipitate in high concentration of organic solvent.

[COSMOSIL SL-II] Wash with methanol, tetrahydrofuran or ethanol.

[Fullerene columns] Wash with *o*-dichlorobenzene, *1,2,4*-trichlorobenzene.

[COSMOSIL Sugar-D/NH_2/HILIC columns] Wash with 50/50 (v/v) acetonitrile/water for NH2-MS and 100% water for Sugar-D and HILIC columns.

Prevention : (a) Choose appropriate pretreatment for each sample. For more information, please see page 193, Technical Information 4.

(b) We also recommend using guard columns, please see page 199, Technical Information 6.

Attention
- When wash columns, do not connect column outflow end to the detector. Let the solvent flow into waste.
- Excessive washing may deteriorate the performance of columns.
- Do not use strongly alkaline solution (more than pH 7.5) or strongly acidic solution (less than pH 1.5) for silica-base packing material.
- Store columns in manufacturer recommended storage solvent after washing.
- If the column performance does not improve after washing, replace the column.

(Case 4) Graduate pressure increase over time

Casue 1 : Contamination of column due to normal long-term use.

Prevention : Wash the column like (Case 3).

Casue 2 : Column damage due to normal long-term use.

Prevention : Replace the columns.

No improvement in performance after washing.

When the column performance has not improved after washing, we recommend replacing the column to lessen pressure burden on the instrument. You could continue to use the column if peak shape is acceptable and the maximum pressure is less than 20 MPa.

4. Sample Pretreatment for HPLC

Pretreatment before HPLC analysis is often required for samples of low concentration or samples containing analytical contaminants. It improves reproducibility and sensitivity in analysis, and protects HPLC columns. The pretreatment methods are different according to the each sample. The followings are examples of different pretreatments.

1) Filtration

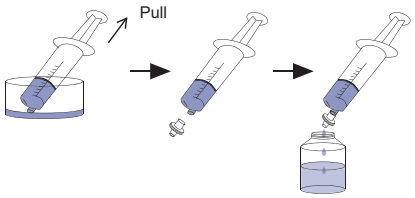
Filtration is a common method used for separating solids from liquids. It extends a column's life by minimizing column damages from solid contaminants such as particles, sediments and colloid substances. It also improves reproducibility of analytical data. We offer both syringe-type and spin-type filters for sample filtration.

	Syringe Filter	Centrifugal Filter
Product	Cosmonice Filter	Cosmospin Filter
Configuration		
Usage	Easy to use Just attach a filter on top of a syringe	Easy to use by centrifugation
Type	W (aqueous system) S (solvent system)	Pore diameter: 0.2 µm Pore diameter: 0.45 µm
Required Equipment	Syringe, Sample Bottle	Centrifuge
Page	Page 80	Page 80

Cosmonice Filter

How to use :

1. Fill a syringe with the sample you want to filter.
2. Attach a Cosmonice filter to the syringe.
3. Push the syringe plunger to filter the sample.
4. Analyze the filtered sample by HPLC.

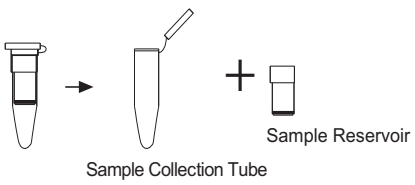


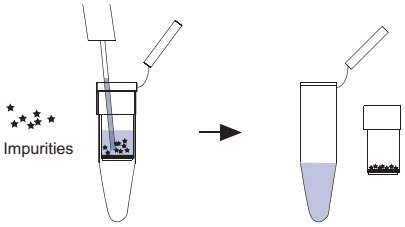
Cosmospin Filter

Components : Sample Reservoir
Sample Collection Tube

How to use :

1. Insert a Cosmospin sample reservoir into a Cosmospin sample collection tube.
2. Add a sample into the Cosmopsin sample reservoir.
3. Close the sample collection tube cap and centrifuge.
4. Remove the sample reservoir and collect the filtered sample in the sample collection tube.
5. Analyze the filtered sample by HPLC.

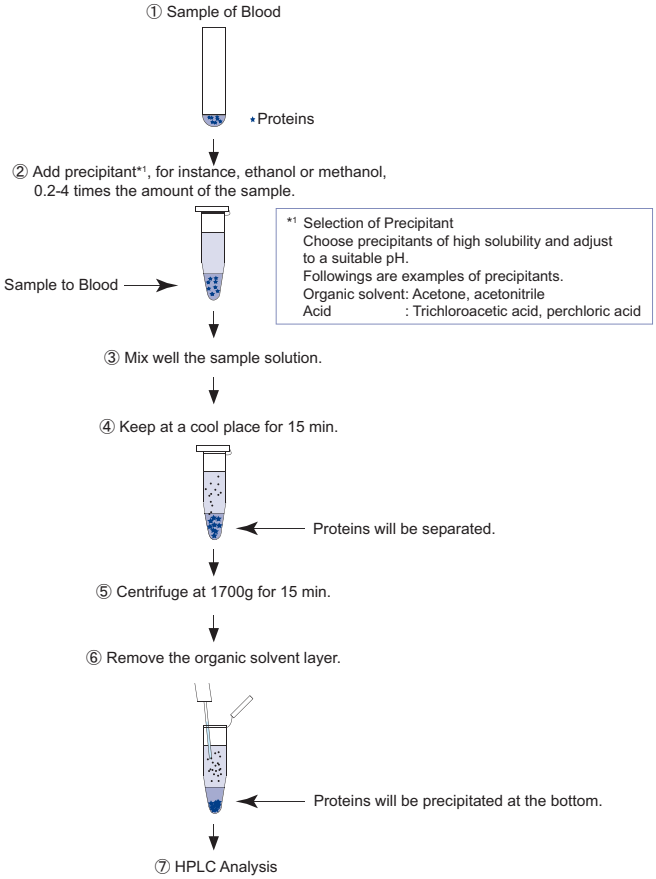




2) Protein Precipitation

Protein precipitation is commonly used to remove proteins in samples for downstream analysis. For example, when analyzing drug concentration in blood samples, proteins have to be removed first. Otherwise, proteins may be adsorbed in columns and interfere with the analysis. Common methods for protein recipitation include salting out, isoelectric point precipitation and precipitation with organic solvents. The following shows a general procedure for protein precipitation with organic solvents.

Procedure for Protein Precipitation :

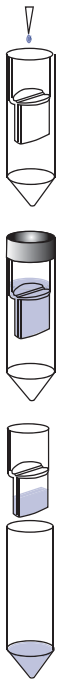


3) Ultrafiltration

Ultrafiltration is a method to concentrate proteins or other macromolecules through a semipermeable membrane with defined pores. Ultrafiltration is applicable for sample desalting, concentrating proteins from dilute solution such as urine samples, or deproteinizing samples with high protein concentration (e.g., blood serum or plasma). Following is a general procedure for ultrafiltration.

Procedure for ultrafiltration :

Procedure for Ultrafiltration :



① Choose an appropriate ultrafiltration tube (filter device). Add sample to the sample reservoir. Close the cap of the filter unit.

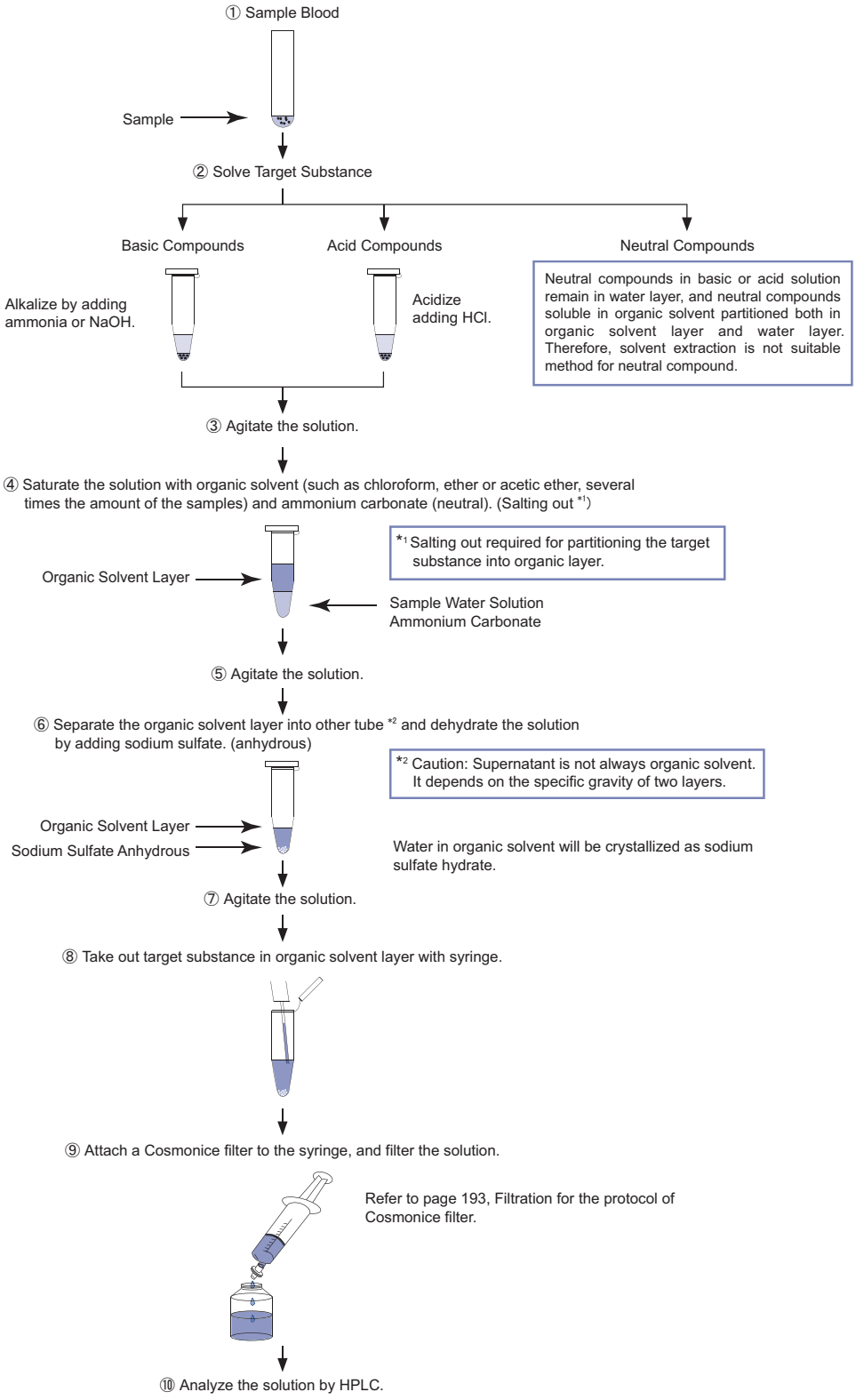
② Centrifuge according to filter device manufasturer's recommendation. Proteins will remain at the filtration membrane. Concentrated protein solution can be recovered from the sample reserver. Filtered solution is in the collection tube.

③ Analyze the filtered solution and/or the concentrated protein solution by HPLC.

4) Solvent Extraction Method

Solvent extraction is a method to separate compounds due to their unequal solubility in two immiscible liquid phases, usually water and an organic solvent. The method is used to concentrate highly hydrophobic compounds, and consequently increase analytical sensitivity. A buffer solution is added to sample to optimize the pH and target substance is then extracted by an organic solvent such as ether and chloroform. However, when target substance is ombined with proteins, solvent extraction may not work well.

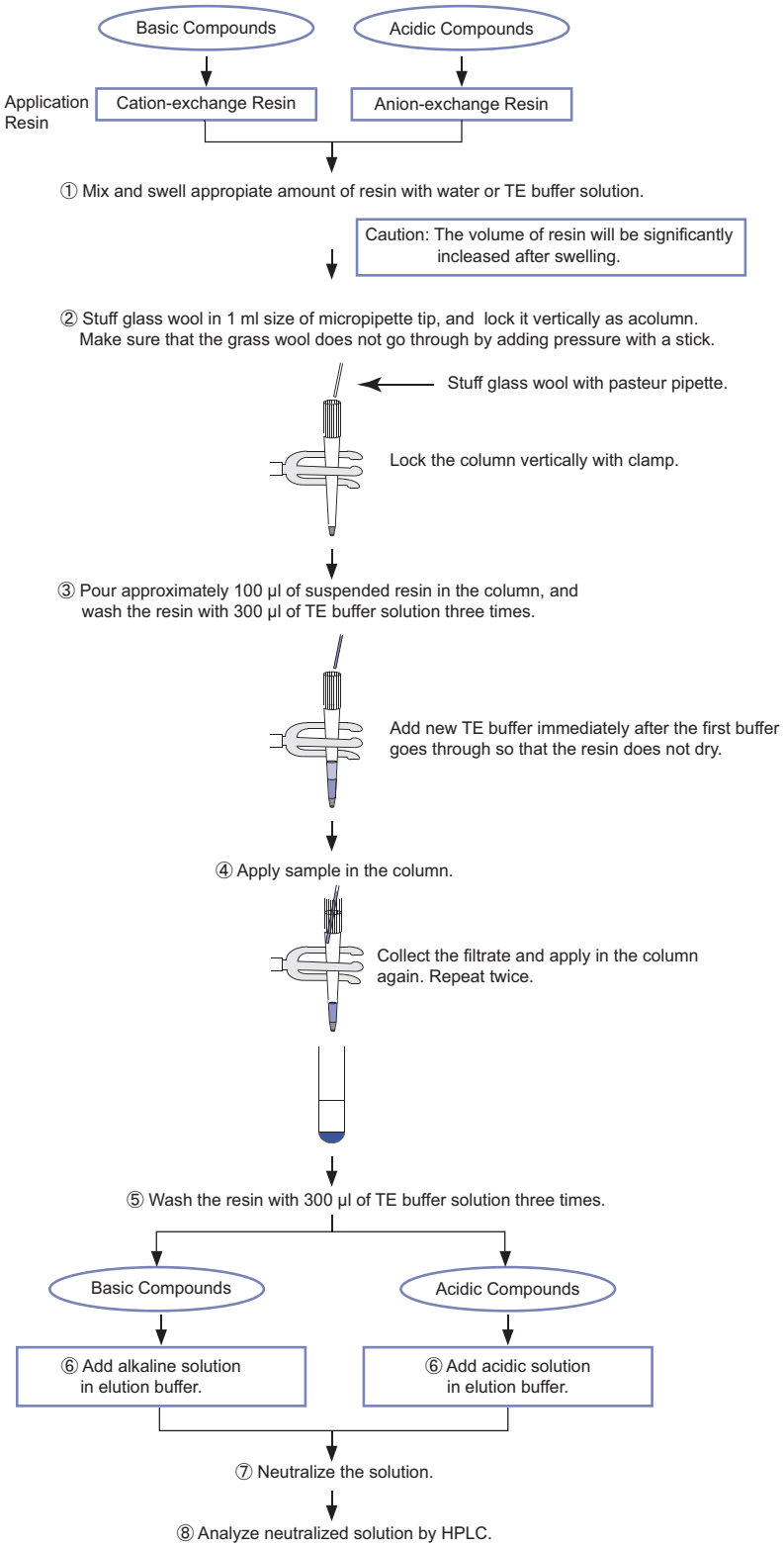
Procedure for Solvent Extraction Method :



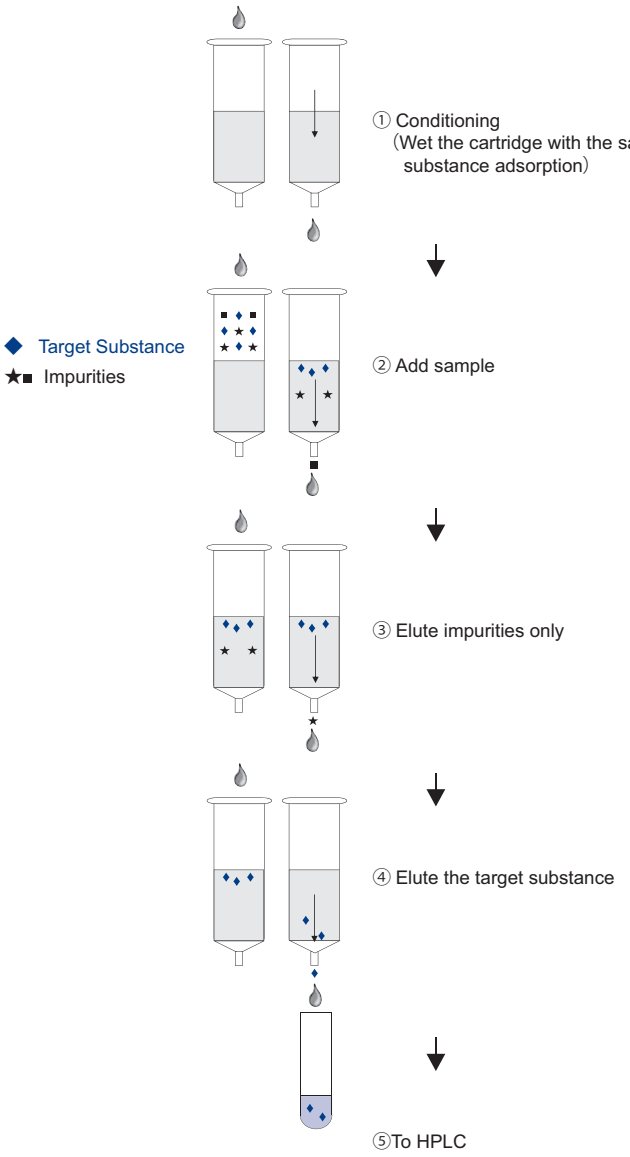
5) Ion Exchange

Pretreatment by ion-exchange resin may be effective for samples that the solvent extraction method cannot be adapted due to its emulsification. A preliminary experiment may be required for the selection of resin and experimental conditions. For example, a negatively charged compound is strongly adsorbed on an anion-exchange resin such as DEAE cellulose resin. Therefore, the target compound is collected by increasing salt concentration of buffer solution or adjusting pH of elution buffer after washing off other weakly adsorbed undesired substances.

Procedure for Ion Exchange :



6) Solid Phase Extraction



5. Baseline Noise in Gradient Elution

In gradient analysis, incomplete mixing of mobile phases or impurities in water of mobile phase can cause baseline noise. In the former case, it can be improved by using a proper mixer before injector (Baseline 1→2). In the latter case, it can be improved by using a pre-column. Impurities in water are adsorbed on the pre-column (Baseline 2→3). COSMOSIL 5C$_{18}$-AR-II 4.6 mm I.D. x 10 mm or 10 mm I.D. x 20 mm as a pre-column.

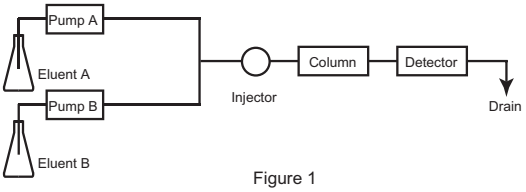
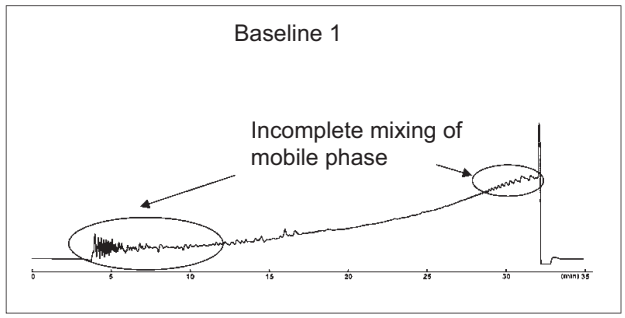


Figure 1



+ Mixer

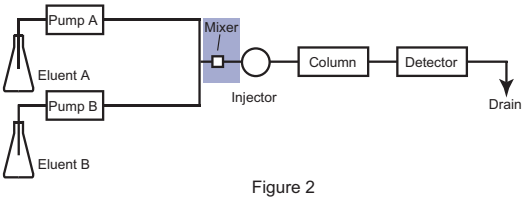
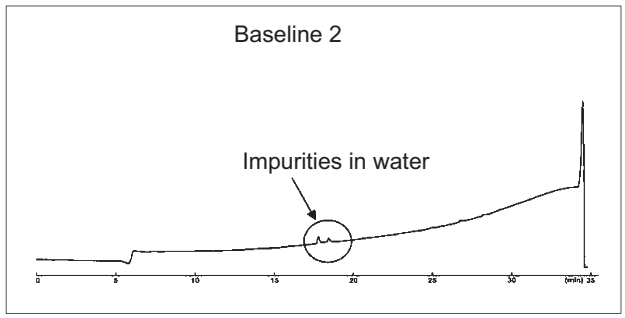


Figure 2



+ Pre-column

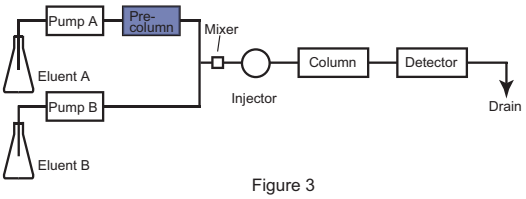
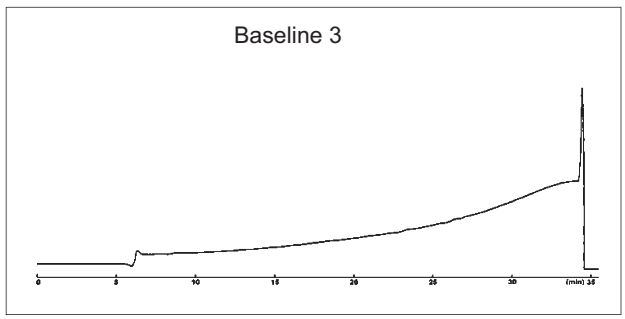


Figure 3



Column	COSMOSIL 5C$_{18}$-AR-300 4.6 mm I.D. x 150 mm
Precolumn	COSMOSIL 5C$_{18}$-AR-II 4.6 mm I.D. x 10 mm
Mobile phase	A: 0.1% TFA containing water
	B: 0.1% TFA containing 95% acetonitrile
	B: 0% → 100%/30 min liner gradient
Flow rate	1.0 ml/min
Temperature	30℃
Detection	UV 220nm

6. Effect of Guard Column

Introduction

The use of guard columns to protect both analytical and preparative columns is highly recommended. COSMOSIL guard columns are packed with the identical packing materials as in analytical and preparative columns. As the result, COSMOSIL guard columns do not affect the performance of the main column.

Selection of Guard Column

Use guard column with the identical packing materials as in the analytical and preparative columns. For guard column size, use the same or smaller inner diameter, and short column length (10–50 mm). For more information on the product code or size, please refer to the respective pages of each column.

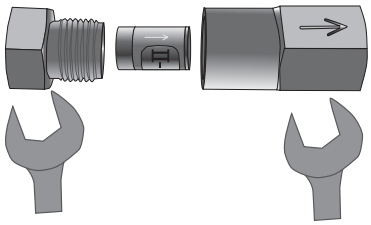
(e.g., Main column 5C$_{18}$-MS-II (20 mm I.D. x 250 mm) → Guard column 5C$_{18}$-MS-II (10 mm I.D. x 20 mm)

Guard Cartridge

For common size columns (4.6 mm I.D. x 10 mm), guard columns and reasonably priced guard cartridges are both available. Guard cartridges are disposable guard columns with identical packing materials as in analytical columns. When using guard cartridges, a COSMOSIL guard cartridge holder (Product No. 38009-79) is required. The holder is reusable.

COSMOSIL Guard Cartridge Holder (Left)
Guard Cartridge (Right)

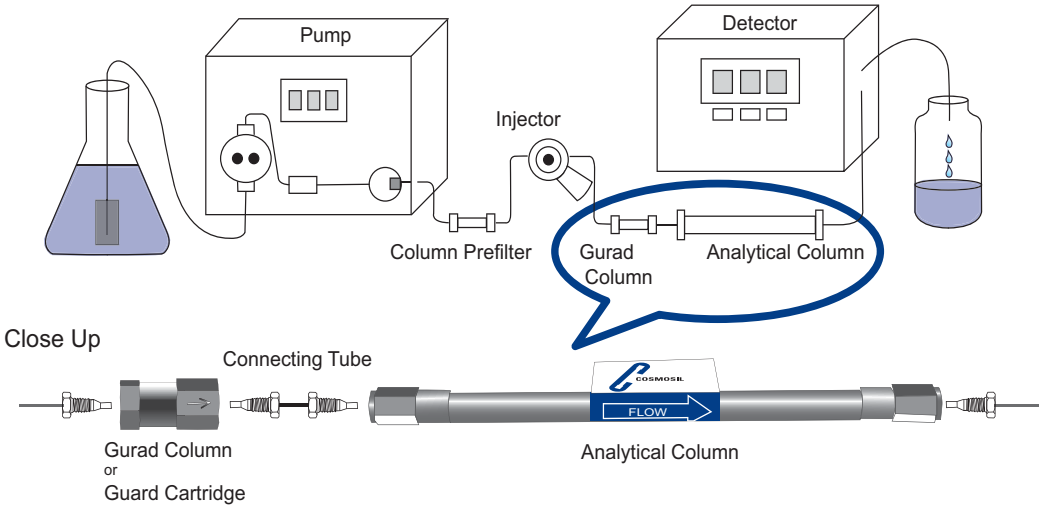
Structure of Cartridge



For more information on how to use the guard cartridge, see the attached instruction manual.

Connection to Guard Column

Use COSMOSIL column connecting tube.(Product No. 37843-69) Please refer to page 87 for more information.



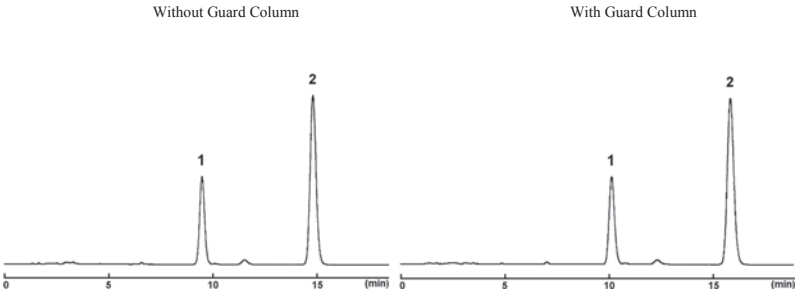
Example of Using Guard Column

The following chromatograms show the use of a COSMOSIL 5C18-MS-II analytical column (4.6 mm I.D. × 150 mm) and the same column connected with its guard column (4.6 mm I.D. × 10 mm). There is no difference in separation characteristics since the packing material is identical in both the guard column and the main column.

Using Guard Column

Column: $5C_{18}$-MS-II
Column size: (Analytical Column) 4.6mmI.D.-150mm
(Guard column) 4.6mmI.D.-10mm
Mobile phase: Methanol/ H_2O = 70/30
Flow rate: 1.0 ml/min
Temperature: 30°C
Detection: UV254nm

Sample: 1; Betamethasone 17-Valerate (0.25 μg)
2; Isoamyl Benzoate (2.5 μg)



NACALAI TESQUE, INC

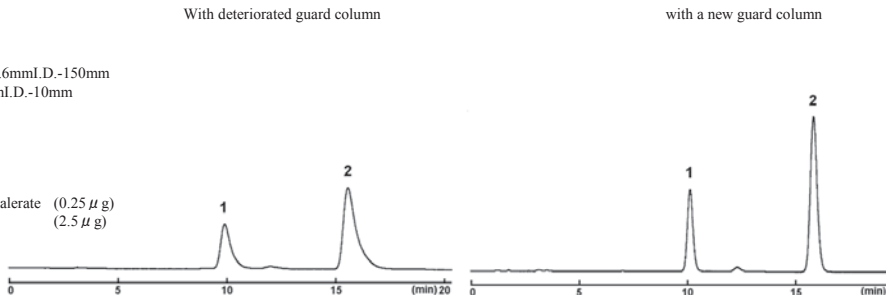
Replacing Guard Column

The following chromatograms show the peak shapes recovered by replacing a deteriorated guard column with a new one. hen pressure increases, ghost peak appears, or base line shifts, promptly replace the guard column with a new one. Continued use of a deteriorated guard column can result in premature deterioration of the main column.

Replacing Guard Column

Column: $5C_{18}$-MS-II
Column size: (Analytical Column) 4.6mmI.D.-150mm
(Guard column) 4.6mmI.D.-10mm
Mobile phase: Methanol/ H_2O = 70/30
Flow rate: 1.0 ml/min
Temperature: 30°C
Detection: UV254nm

Sample: 1; Betamethasone 17-Valerate (0.25 μg)
2; Isoamyl Benzoate (2.5 μg)



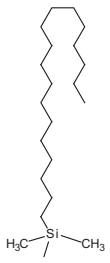
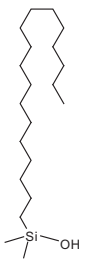
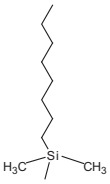
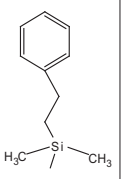
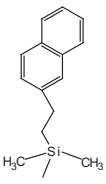
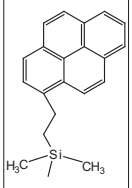
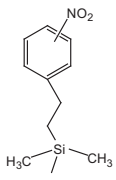
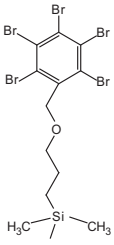
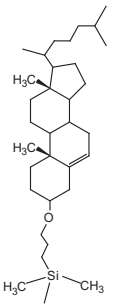
NACALAI TESQUE, INC

7. Selectivity of Packing Materials in Reversed Phase Liquid Chromatography

Introduction

Reversed phase chromatography is the most commonly used method of HPLC, because of the high theoretical plate number, excellent separation characteristics, reproducibility, and ease of use. Columns packed with octadecyl group bonded type silica gel (C_{18}, ODS) are the most widely used reversed phase chromatography. However, C_{18} columns provide insufficient separation for compounds similar in hydrophobicity because the main separation mechanism of C_{18} column is based on hydrophobic interaction. It may improve separation of compounds with similar hydrophobicity by using longer columns, changing mobile phases or changing temperature. However, in many cases, it is probably most effective to use different packing materials which retain compounds base on a secondary interaction in addition to hydrophobic interaction. At Nacalai, we offer a variety of COSMOSIL reversed phase packing materials. Summary of these packing materials and their respective retention mechanism are in Table 1. Retention of compounds in each stationary phase depends on summation of the interactions. Therefore, comprehension of each interaction leads to selection of an appropriate column.

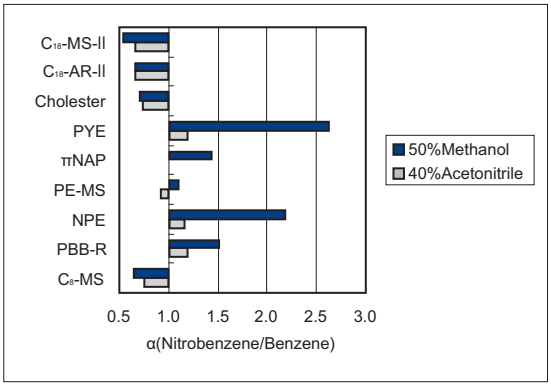
Table1. Stationary phase and interaction of packing materials

Packing Material	C_{18}-MS-II	C_{18}-AR-II	C_8-MS	PE-MS	πNAP	PYE	NPE	PBB-R	Cholester
Silica Gel	High Purity Porous Spherical Silica								
Average Particle Size	5 µm								
Average Pore Size	approx. 120 Å								
Specific Surface Area	approx. 300 m^2/g								
Bonded Phase									
	Octadecyl Group	Octadecyl Group	Octyl Group	Phenylethyl Group	Naphthylethyl Group	Pyrenylethyl Group	Nitrophenylethyl Group	Pentabromobenzyl Group	Cholesteryl Group
Bonding Type	Monomeric	Polymeric	Monomeric	Monomeric	Monomeric	Monomeric	Monomeric	Monomeric	Monomeric
Main Interaction	Hydrophobic Interaction	Hydrophobic Interaction	Hydrophobic Interaction	Hydrophobic Interaction	Hydrophobic Interaction	Hydrophobic Interaction π−π Interaction Dispersion Force Charge-transfer Interaction	Hydrophobic Interaction π−π Interaction Dipole-dipole Interaction	Hydrophobic Interaction Dispersion Force Interaction	Hydrophobic Interaction Molecular Shape Selectivity
End-capping Treatment	Near-perfect Treatment								
Carbon Load	approx. 16%	approx. 17%	approx. 10%	approx. 10%	approx. 11%	approx. 18%	approx. 9%	approx. 8%	approx. 20%

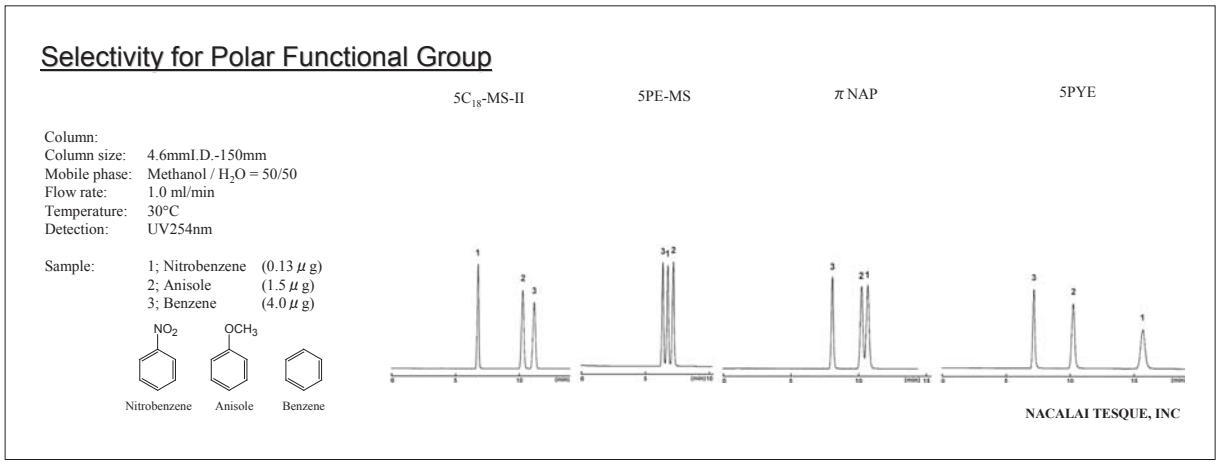
1) Selectivity for Polar Functional Group

Selectivity

Selectivity for polar functional group is evaluated based on the separation of benzene, nitrobenzene, which has a nitro group, and anisole, which has a methoxy group. The chromatograms below show separation of the three compounds on four COSMOSIL columns : C_{18}-MS-II, PE-MS, πNAP and PYE. Elution order on the C_{18} column is as following : nitrobenzene, anisole and benzene. Elution orders on the aromatic columns are reversed. Separation on the C_{18} column is based on hydrophobic interaction only. On the other hand, the packing materials on the other three columns have aromatic rings and reverse the elution order by π–π interaction.



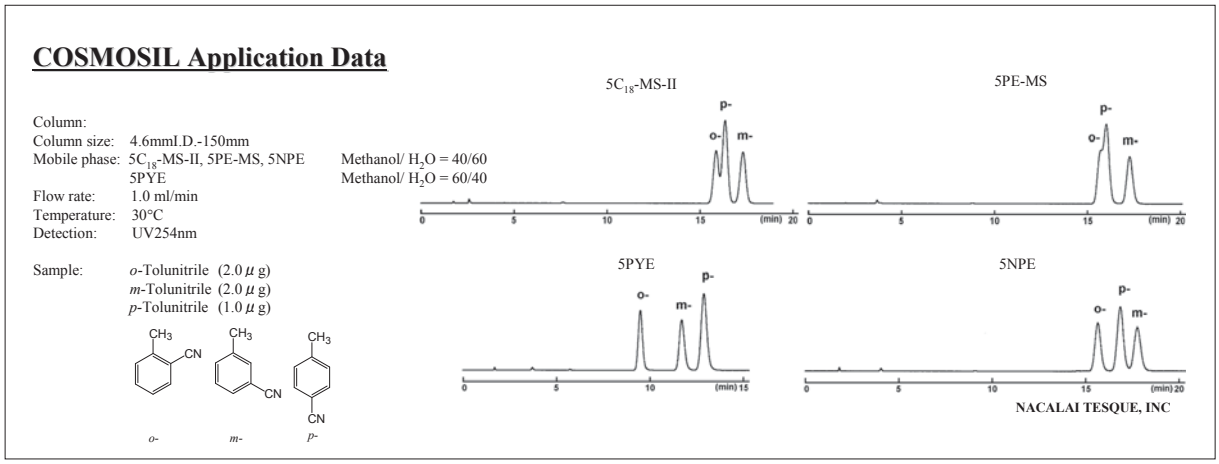
The graph of selectivity for polar functional group is shown below. Among nine COSMOSIL columns, PYE and NPE columns have the highest selectivity factors for polar groups. As to mobile phases, methanol is more effective than acetonitrile for separation using π–π interaction.



Application

- Separation of Tolunitrile Position Isomers

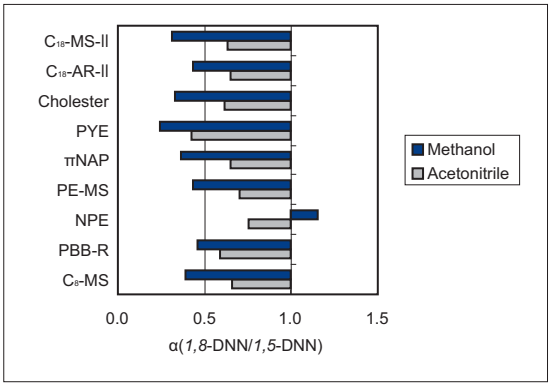
Tolunitriles have three position isomers. It is difficult to separate ortho and para isomers by C_{18} or PE–MS column because of lack of poor π–π interaction. On the other hand, the isomers are well separated on PYE or NPE column which has strong π–π interaction.



2) Selectivity for Dipole

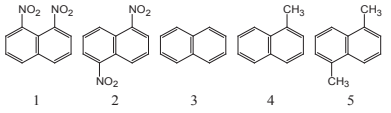
Selectivity

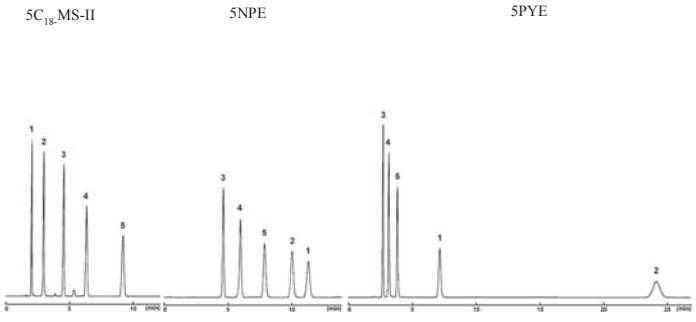
Selectivity for dipole is evaluated based on the separation of *1,5*–dinitronaphthalene and *1,8*–dinitronaphthalene. Dinitronaphthalenes (peak 1 and 2) were strongly retained on PYE and NPE because of π–π interaction compared with dimethylnaphthalenes. However, there is a slight difference between these two columns. While *1,5*–dinitronaphthalene (peak 2) was preferentially retained on PYE, *1,8*–dinitronaphthalene (peak 1) was retained longer on NPE. The results with NPE indicate the presence of strong dipole–dipole interaction. The two nitro group dipoles in *1,8*–dinitronaphthalene are aligned for a much greater dipolar coupling with the bonded nitrophenyl group in NPE than *1,5*–dinitronaphthalene.



Selectivity for Dipole

Column size: 4.6mmI.D.-150mm
Mobile phase: $5C_{18}$-MS-II Methanol / H_2O = 80/20
NPE Methanol / H_2O = 70/30
PYE Methanol / H_2O = 90/10
Flow rate: 1.0 ml/min
Temperature: 30°C
Detection: UV254nm
Sample: 1; *1,8*-Dinitronaphthalene(*1,8*-DNN) (0.21 μg)
2; *1,5*-Dinitronaphthalene(*1,5*-DNN) (0.11 μg)
3; Naphthalene (0.25 μg)
4; *1*-Methylnaphthalene (0.35 μg)
5; *1,5*-Dimethylnaphthalene (0.42 μg)





Application

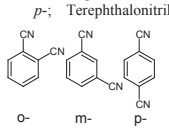
● Separation of Phthalonitrile Position Isomers

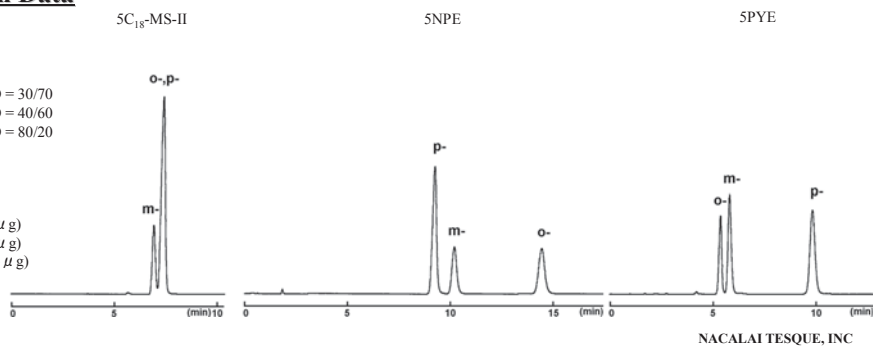
Phthalonitriles have three position isomers. NPE or PYE completely separates these compounds due to π–π interaction. Furthermore, NPE strongly retains o–phthalonitrile due to dipole–dipole interaction.

COSMOSIL Application Data

Column:
Column size: 4.6mmI.D.-150mm
Mobile phase: $5C_{18}$-MS-II Methanol/ H_2O = 30/70
5NPE Methanol/ H_2O = 40/60
5PYE Methanol/ H_2O = 80/20
Flow rate: 1.0 ml/min
Temperature: 30°C
Detection: UV254nm

Sample: *o*-; Phthalonitrile (0.3 μg)
m-; Isophthalonitrile (3.0 μg)
p-; Terephthalonitrile (0.15 μg)

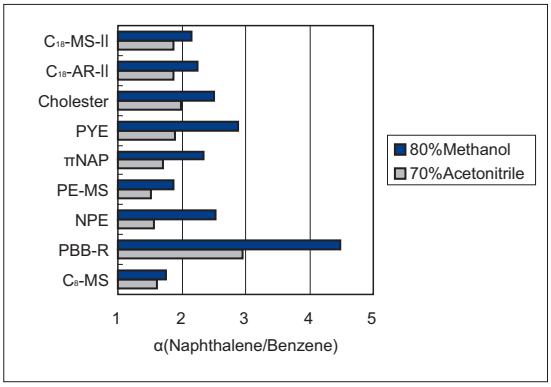


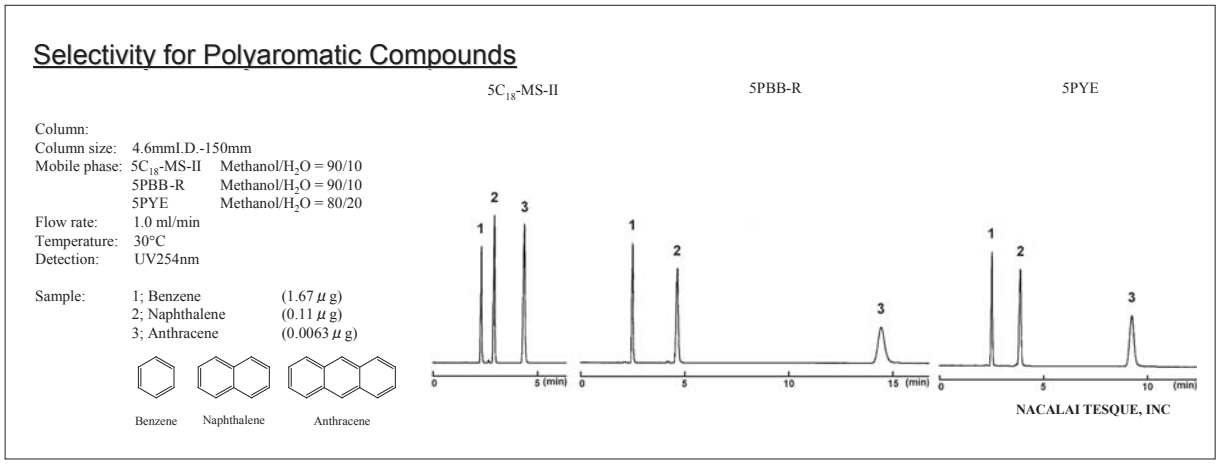


3) Selectivity for Polyaromatic Compounds

Selectivity

Selectivity for polyaromatic compounds is evaluated based on the separation of benzene, naphthalene and anthracene. The elution orders in all columns are the same : benzene, naphthalene and anthracene. Retention increases in all columns with increasing number of aromatic rings. In addition, highly dispersive packing materials such as PBB and PYE show much stronger retention for polyaromatic compounds due to dispersion interaction.

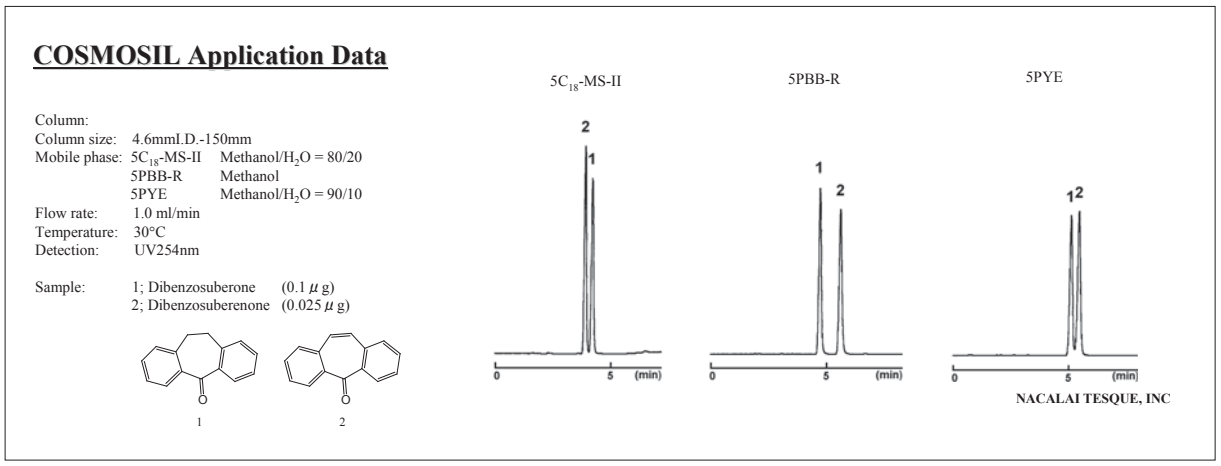




Application

● Separation of Dibenzosuberone and Dibenzosuberenone

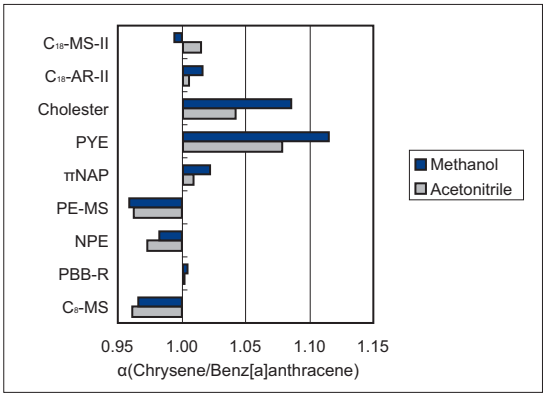
C_{18} retains dibenzosuberone (peak 1) longer than dibenzosuberenone (peak 2). On the other hand, PBB–R and PYE retain dibenzosuberenone (peak 2), which has a π–electron conjugated system, longer than dibenzosuberone (peak 1).

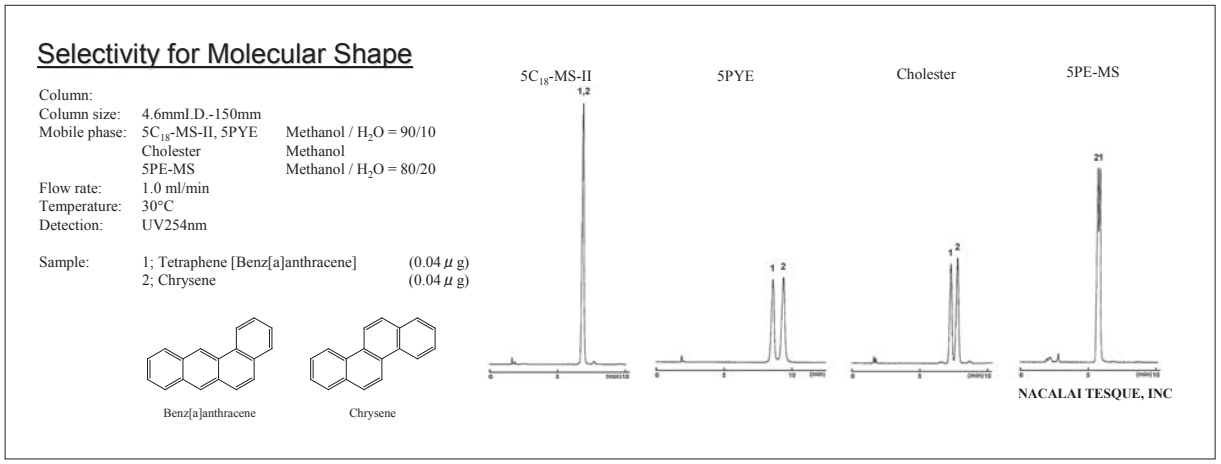


4) Selectivity for Molecular Shape

Selectivity

Selectivity for Molecular Shape is evaluated based on the separation of chrysene and benz[a]anthracene. The isomers of two polyaromatic hydrocarbons, which consist of four benzene rings, are difficult to separate because of the similar hydrophobicity or aromaticity. However, PYE and Cholester columns, which recognize molecular shape, enable them to separate chrysene and benz[a]anthracene.

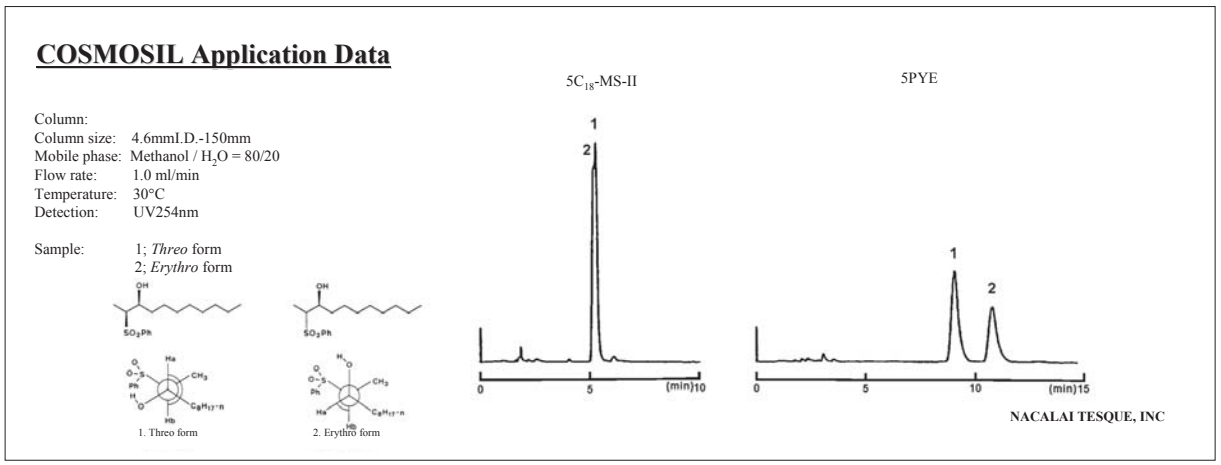




Application

● Separation of Diastereomers (*threo*- and *erythro*-)

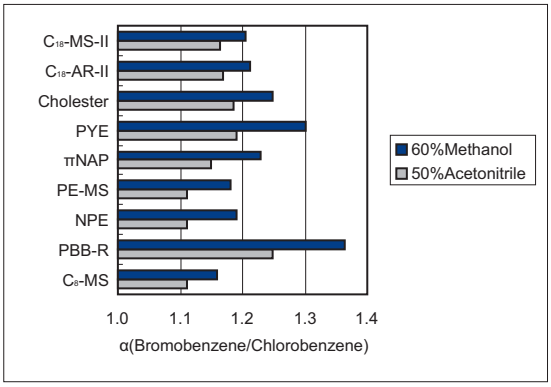
C_{18} cannot separate the threo and erythro forms. On the other hand, PYE retains the planar erythro form longer than the threo form.



5) Selectivity for Halides

Selectivity

Selectivity for halides is evaluated based on the separation of chlorobenzene and bromobenzene. PBB-R shows the highest selectivity factor due to dispersion interaction of the five bromine atoms.

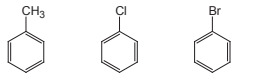


Application

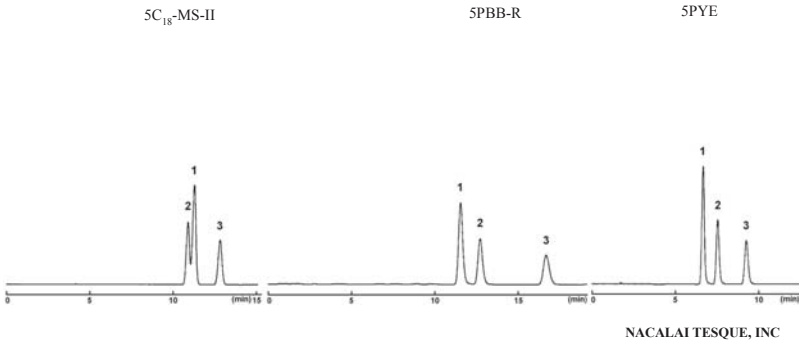
Selectivity for Halide

Column:
Column size: 4.6mmI.D.-150mm
Mobile phase: Methanol/ H_2O = 60/40
Flow rate: 1.0 ml/min
Temperature: 30°C
Detection: UV254nm

Sample: 1; Toluene (3.3 μg)
2; Chlorobenzene (3.3 μg)
3; Bromobenzene (3.3 μg)



Toluene Chlorobenzene Bromobenzene



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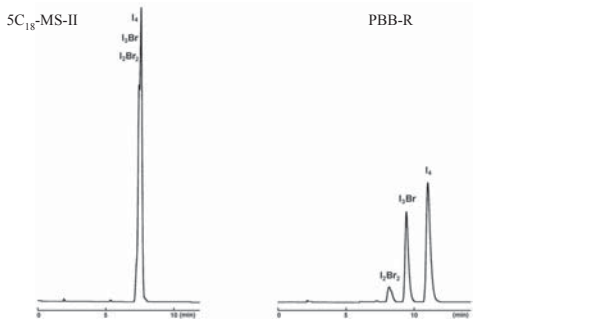
● Separation of Halogen Exchange Reaction Products

PYE and PBB-R retain dispersed iodine atom longer than bromine atom. As a result, PYE and PBB-R can separate the complicated bromine and iodine compounds that C18 cannot separate.

COSMOSIL Application Data

Column:
Column size: 4.6mmI.D.-150mm
Mobile phase: 5C18-MS-II Methanol/ H_2O = 90/10
PBB-R Methanol
Flow rate: 1.0 ml/min
Temperature: 30°C
Detection: UV254nm

Sample:



Sample courtesy of Dr.H.Yamamoto,RIKEN

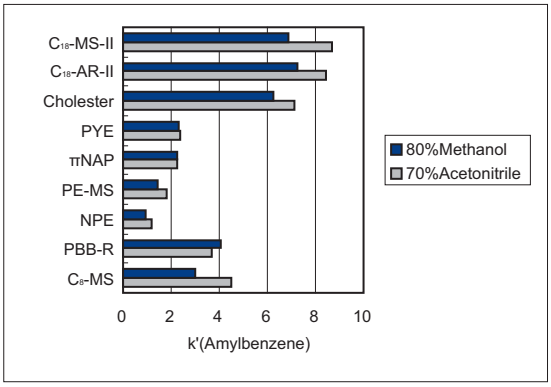
NACALAI TESQUE, INC

AP-1029

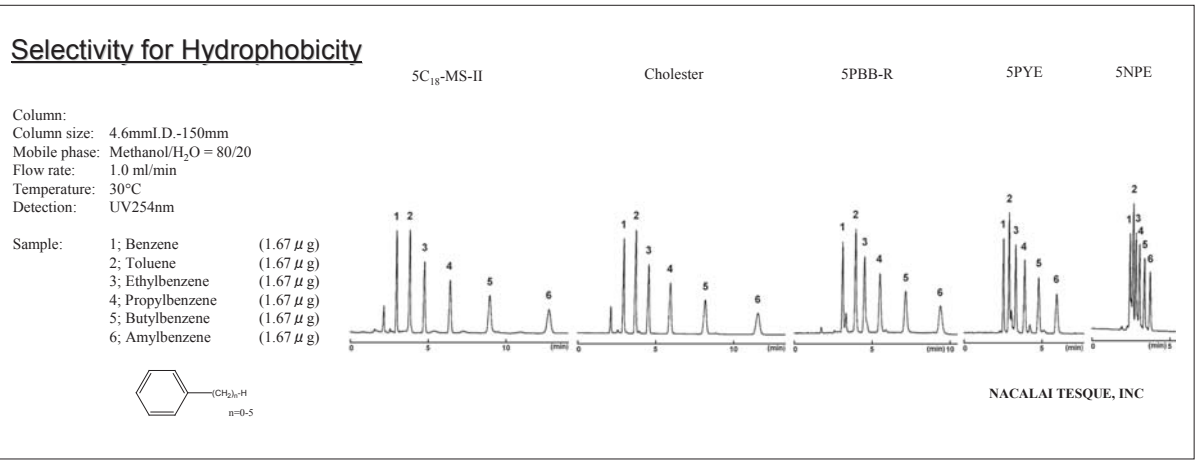
6) Selectivity for Hydrophobicity

Selectivity

Selectivity for hydrophobicity is evaluated based on the separation of alkylbenzenes. Two C_{18} and Cholester show similar high selectivity for hydrophobicity. Other columns show less hydrophobic selectivity than C_{18}.

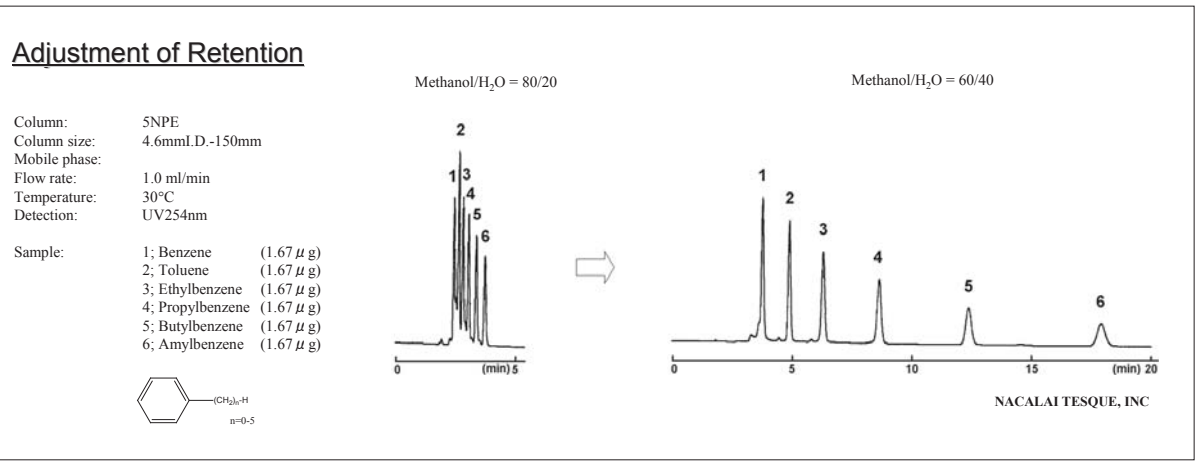


Selectivity for Hydrophobicity



Lower concentration of organic solvent in mobile phase leads to much retention in reversed phase chromatography. In case of NPE, when methanol concentration is reduced to 60%, the retention times increase to those similar to C_{18} with 80% methanol.

Adjustment of Retention



8. Methods in Developing Mobile Phase Condition for C_{18} Column

Introduction

In reversed phase HPLC, octadecyl group bonded silica columns (C_{18}, ODS) are the most widely employed. A proper mobile phase condition for C_{18} columns can be achieved by referring to publications, application notes from manufactures and your own experiences. This section shows traditional methods of developing mobile phase condition. The following columns are used as examples because of their popularity.

Packing material : COSMOSIL 5C_{18}-MS-II, COSMOSIL 5C_{18}-AR-II
Column size (I.D. x length) : 4.6 mm I.D x 150 mm

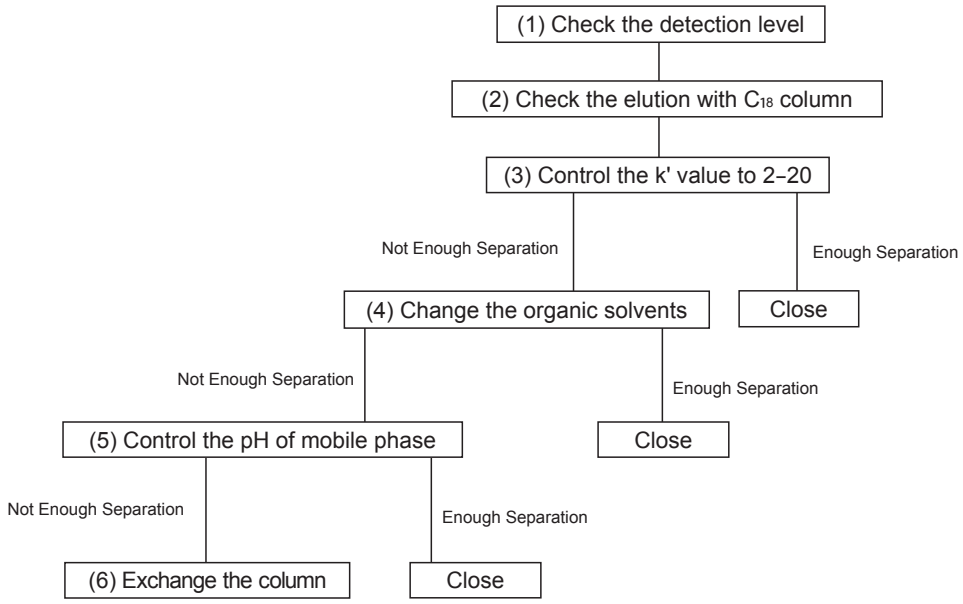
Methods for Developing Mobile Phase Conditions

In the Isocratic method, the mobile phase composition remains constant throughout the run. In the gradient method, the mobile phase composition changes. Each method needs strict preparation of the mobile phase and control of the column temperature to achieve good separation.

● Isocratic Method

Methods for developing mobile phase condition generally proceed as follows. First, elute the samples with strong solvents to check whether the samples can be detected. Then, separate the samples by controlling the retention time through changing the mobile phase condition. Increasing the concentration of strong elution efficiency solvents results in shorter retention time and decreasing in concentration of them results in longer retention time. If your samples are ionizable, such as acid and amine group, pH control with buffers is highly advisable.. The ionization control method or ion-pair chromatography is used to increase the retention of ionized samples. The method uses ion-pair reagents (e.g., alkyl benzene sulfonate for basic compounds, quaternaty ammonium for acidic compounds) into in the mobile phase to form ion pairs with samples.

(e.g.,) Procedures for Basic Condition Setting



1. Check the detection of samples with strong elution solvents. In this step, check the detection by connecting the injector directly to the detector without a column.
2. Consult references and carbon numbers and check the elution with C_{18} column using aqueous mobile phase with methanol.
3. Control the k' value to 2–20 by changing the amount of the methanol in the mobile phase.
4. If the separation is not enough, change the methanol to acetonitrile or add tetrahydrofuran to change the selectivity.
5. If tailing peaks occur for basic compounds, control the pH by adding buffers to the mobile phase.
6. If separation is not satisfactory after step 5, change the column to other C_{18} columns or columns with different stationary phases such as alkyl–based, aromatic–based and others.

● Gradient Method

Gradient method changes organic solvent composition continuously in the mobile phase. It is useful for shortening the separation time of samples with wide range of hydrophobicity and molecular weight, with long elution time, and with great changes in retention time by slight changes in organic solvent composition. It is also useful for large molecule weight compounds like peptides. Gradient method is not compatible with RI detector. Gradient method development is beyond the scope that can be discussed here.

Easy Method to Set the Reversed Phase Condition

Reversed phase chromatography does not have an easy method to set mobile phase conditions, unlike normal phase chromatography mobile phase conditions that can simply be determined by thin layer chromatography. Therefore, the composition of the mobile phase (concentration of organic solvent) is often determined by repeated trial and error. If you know the structure of the analytes, here is a general instruction on configuring an appropriate mobile phase organic solvent concentration.

Suitable concentration of organic solvent for basic chemical skeleton + effects from substituents
= The best organic solvent concentration

● Condition Setting

Select the condition based on retention time of the basic chemical skeleton shown in Figure 1, then adjust for the effect of hetero atom and substituents.

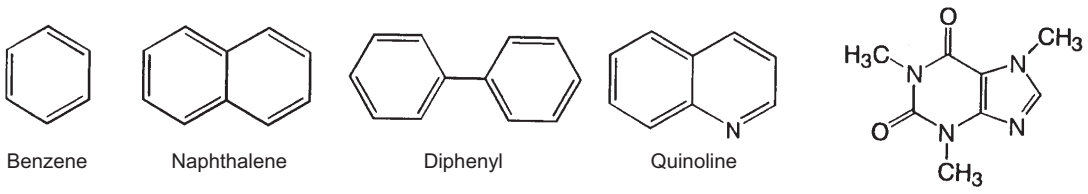


Figure 1. Structures of basic chemical skeletons

1. Select the best concentration of organic solvent with its corrsponding basic chemical skeleton. *Refer Table 1
2. Adjust the concentration of the organic solvent considering effect of hetero atom. *Refer Table 2
3. Adjust the concentration of organic solvent considering the effects from substituents. *Refer Table 3
   - Contain dissociate substituent → 4. Adjust the concentration of the organic solvent considering effects from dissociable substituent. → Complete
   - No dissociate substituent → Complete

1. Choose a compound of similar basic chemical structure as the target sample in Figure 1. Select the best organic solvent concentration.

Table 1. Retention time of the basic chemical skeleton

Basic skeleton	Column	Retention Time Under Different Methanol Concentrations (min)						
		80%	70%	60%	50%	40%	30%	20%
Benzene	5C$_{18}$-MS-II	–	4	7	11	20	–	–
	5C$_{18}$-AR-II	–	4	7	13	23	–	–
Naphthalene	5C$_{18}$-MS-II	5	8	18	–	–	–	–
	5C$_{18}$-AR-II	5	10	22	–	–	–	–
Diphenyl	5C$_{18}$-MS-II	8	13	–	–	–	–	–
	5C$_{18}$-AR-II	7	15	–	–	–	–	–
Quinoline	5C$_{18}$-MS-II	–	–	–	–	6	11	–
	5C$_{18}$-AR-II	–	–	–	–	8	17	–
Caffeine	5C$_{18}$-MS-II	–	–	–	–	–	4	9
	5C$_{18}$-AR-II	–	–	–	–	–	4	9

Column: COSMOSIL 4.6 mm I.D. × 150 mm Flow Rate: 1.0 ml/min Detection: UV 254 nm

2. Adjust the organic solvent concentration considering effect of hetero atom as shown in Table 2.

Table 2. Organic solvent concentration adjustment from hetero rings or polycyclic aromatics

Hetero Rings, polycyclic Aromatics		Sample	$5C_{18}$-MS-II	$5C_{18}$-AR-II
1 of Conjugate Ring		Benzene	+10%	+10%
Heterocyclic Hetero Atom	1 of S	Thiophene	±0%	±0%
	1 of O	Furane	-5%	-5%
	1 of N	Pyridine	-20%	-10%
1 of Carbonyl Group		Quinone	-5%	-5%
1 of Double Bond		-	-5%	-5%

3. Adjust the concentration of organic solvent considering effect from substituents as shown in Table 3.

Table 3. Organic solvent concentration adjustment from substituents

Substituent	Methanol Concentration	
	$5C_{18}$-MS-II	$5C_{18}$-AR-II
-F	0	0
-Cl	+10%	+10%
-Br	+10%	+10%
-I	+20%	+15%
$-CONH_2$	-40%	-40%
$-COCH_3$	-10%	-10%
$-COOCH_3$	0	0
$-OCH_3$	0	0
$-CHCH_2O$	-10%	-10%
-CH2OH	-30%	-30%
-OH	-30%	-30%
$-NO_2$	-10%	-5%
-CN	-20%	-15%
$-NH_2$	-40%	-30%
$-SCH_3$	+10%	+10%

Substituent	Methanol Concentration
-CH2- (Alkyl-chain) MeOH concentration of basic skeleton	
100-90%	+10% (4 of $-CH_2-$)
90-80%	+10% (3 of $-CH_2-$)
80-60%	+10% (2 of $-CH_2-$)
< 60%	+10% (1 of $-CH_2-$)
-Phenyl MeOH concentration of basic skeleton	
100-90%	+5% (1 of – Phenyl)
90-60%	+10% (1 of – Phenyl)
< 60%	+20% (1 of – Phenyl)

Column: COSMOSIL 4.6 mm I.D. × 150 mm

Flow Rate: 1.0ml / min Detection: UV 254 nm

* Effect may shift somewhat by the position of the substituent.

4. Compounds with a dissociative substituent are extremely sensitive toward slight pH change. Maintain consistent mobile phase pH to obtain reproducible data. Table 4 shows the influence of acidic (pH 2) and neutral (pH 7) substituents to the retention.

Table 4. Effect of dissociate substituent to organic solvent

Dissociable Substituent	Change of Methanol Concentration (pH 2)	Change of Methanol Concentration (pH 7)
-COOH	-10~-20%	-30~-40%
$-SO_3H$	-20~-40%	-30~-40%
$-PO_4H_2$	-20%	-50%
$-BO_2H_2$	-20%	-20%
$-NH_2$ (molecular type)	-60%	-10%
$-NH_2$ (cyclic amine)	-50~-60%	-10~-20%
$-NH_2$ (ionic type)	-	-40~-50%

Column :COSMOSIL $5C_{18}$-MS-II, 4.6 mm I.D. x 150 mm
Buffer pH2 :20mmol/l H_3PO_4
pH7 :20mmol/l H_3PO_4/Na_2HP0_4=2/3
Flow Rate :1.0 ml/min
Detection :UV 254 nm

● Example of Condition Setting

Column: COSMOSIL 5C_{18}-MS-II 4.6 mm I.D. × 150 mm

(1) 5-Benzyloxyindole

<Calculation> Basic skeleton Naphthalene like + (hetero ring N)
=70%+ (-20%)
=50%

Substituent (Phenyl) + (-OCH_2- is equal to -OCH_3)
=(+10%) + (+0%)

Basic skeleton + Substituent = 50% + (+10%) = 60%

<Result> 60%Methanol (Methanol:Water=60:40)
Retention time=13.7 min

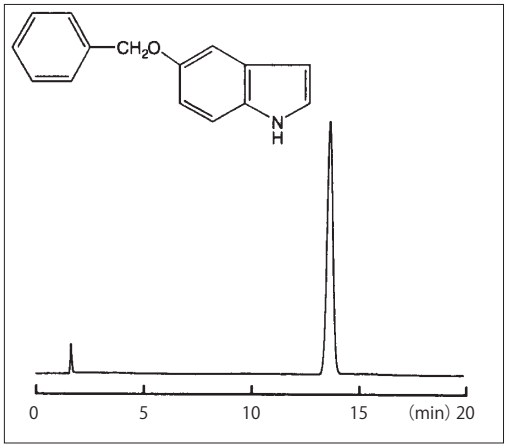


Figure 2. Analysis of 5-Benzyloxyindole

(2) Homovanillic Acid

<Calculation> Basic skeleton Benzene=60%

Nondissociative substituent (-OH) + (-OCH_3) + (-CH_2)
=(-30%) + (0%) + (+10%)
= -20%

Dissociable substituent -COOH = -10~-20% (pH 2)
-30~-40% (pH 7)

Basic skeleton + Substituent = Methanol concentration is
Acid range (pH 2) 30-20%
Neutral range (pH 7) 10-0%

<Result> (pH2) 30%Methanol : Retention time=5.7 min
20%Methanol : Retention time=11.7 min

(pH7) 10%Methanol : Retention time=4.0 min
0%Methanol : Retention time=12.1 min

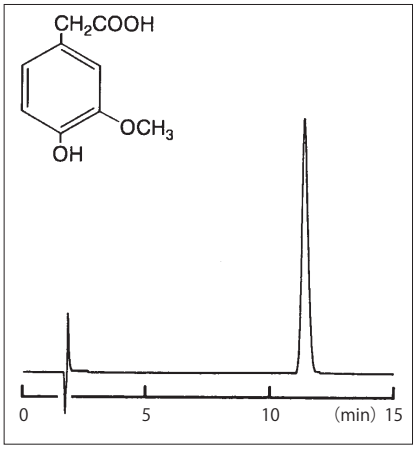


Figure3. Analysis of 20% Methanol (pH 2)

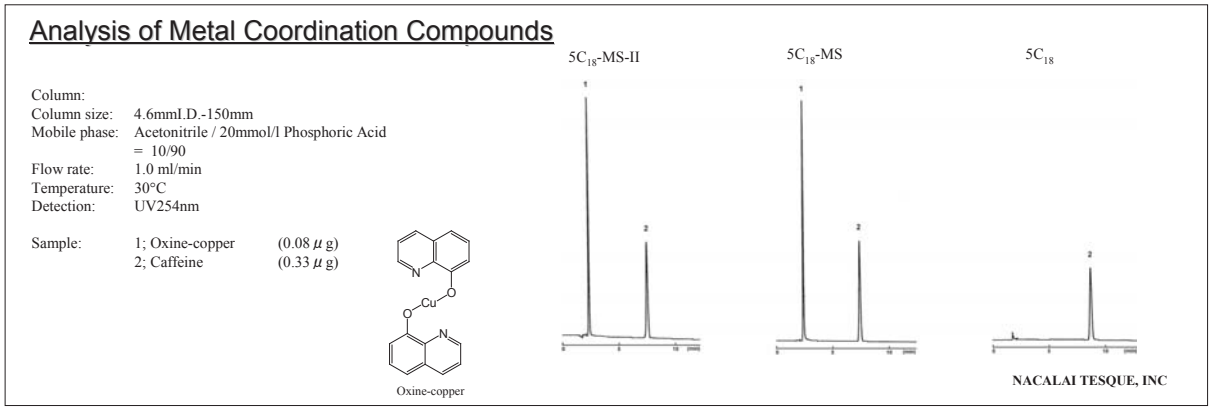
* Actual retention prediction results may have ±10% error in organic solvent concentration calculated.

9. Comparison with Old Type COSMOSIL

1) New Type COSMOSIL (5C$_{18}$-MS-II) vs. Old Type COSMOSIL (5C$_{18}$ and 5C$_{18}$-MS)

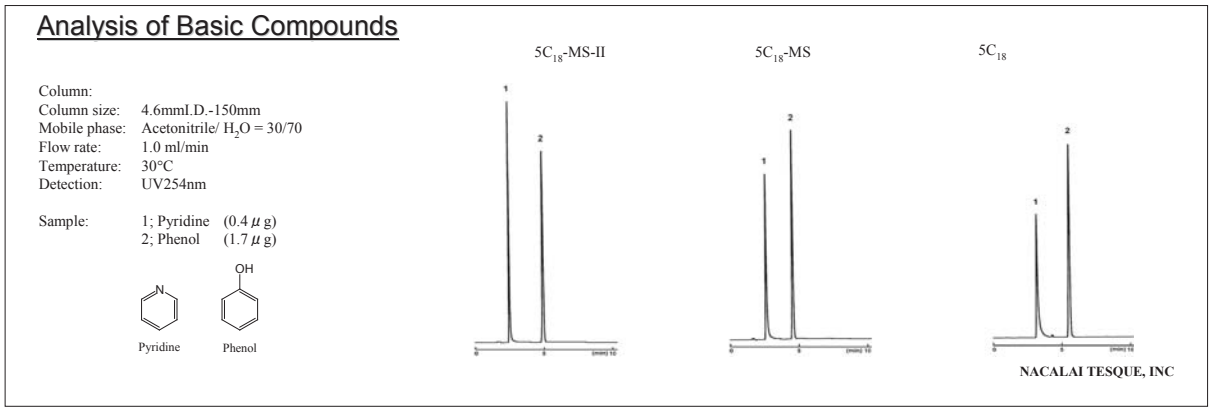
Analysis of Metal Coordination Compounds

The metal coordination compounds, e.g., Oxine–copper, were not eluted from COSMOSIL 5C$_{18}$ because its silica gel contains a high level metal impurities. COSMOSIL 5C$_{18}$–MS or 5C$_{18}$–MS–II can separates the same metal coordination compounds because they are packed with high purity (99.99%) silica gel.



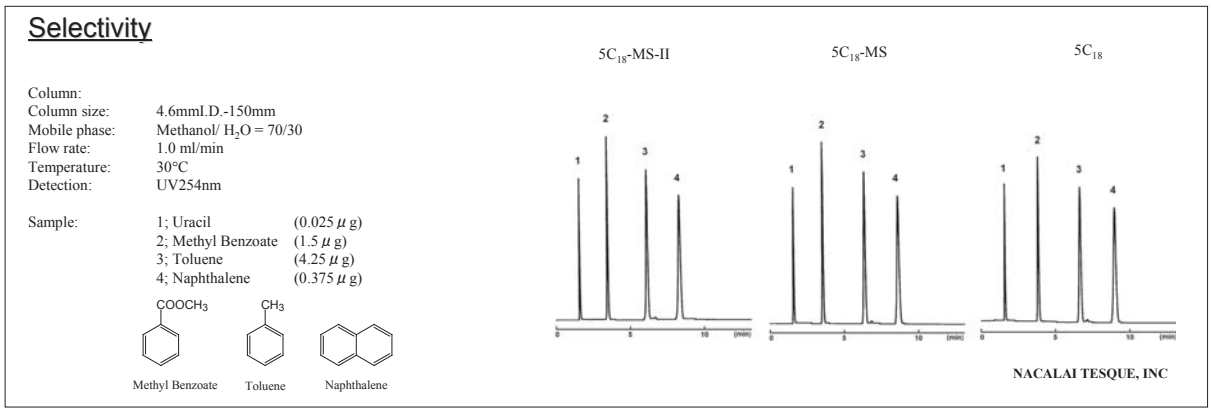
Analysis of Basic Compounds

COSMOSIL 5C$_{18}$–MS–II shows better separation for the basic compounds than COSMOSIL 5C$_{18}$–MS because COSMOSIL 5C$_{18}$–MS–II is treated with improved end–capping.



Selectivity

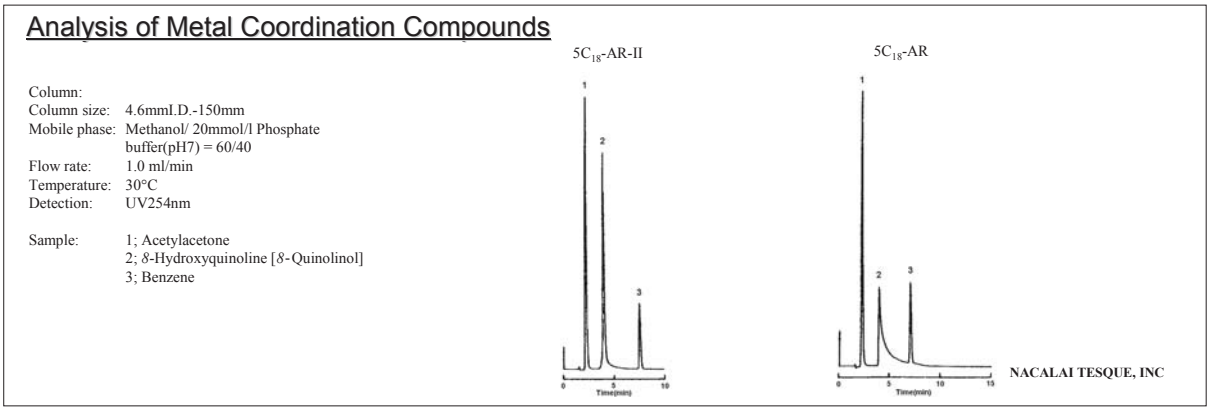
Little difference exists among COSMOSIL 5C$_{18}$, 5C$_{18}$–MS and 5C$_{18}$–MS–II in selectivity. The same analytical condition used for the old type column can be transferred to COSMOSIL 5C$_{18}$–MS–II without any modification.



2) New Type COSMOSIL (5C$_{18}$-AR-II) vs. Old Type COSMOSIL (5C$_{18}$-AR)

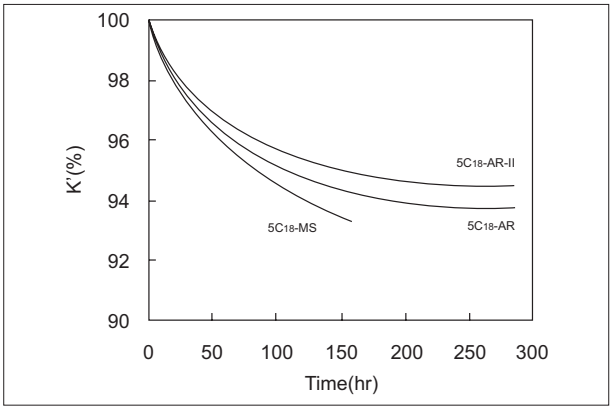
Analysis of Metal Coordination Compounds

COSMOSIL–5C$_{18}$ AR–II shows better separation for the metal coordination compounds e.g., *8*–Quinolinol than COSMOSIL 5C$_{18}$–AR because of the high purity silica gel.



Acid Resistance

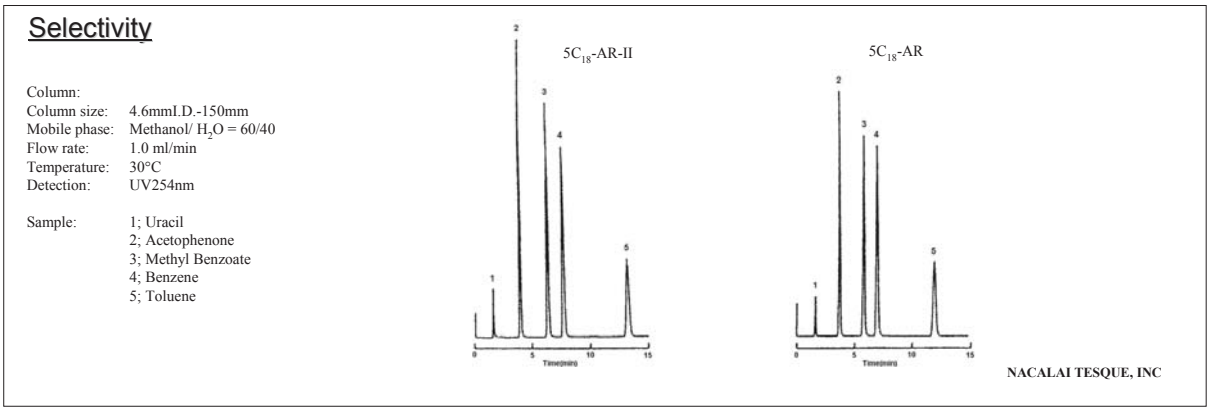
COSMOSIL 5C$_{18}$–AR–II show superior acid resistance to 5C$_{18}$–AR.



Degradation test with 0.1% Trifluoroacetic Acid at 60°C

Selectivity

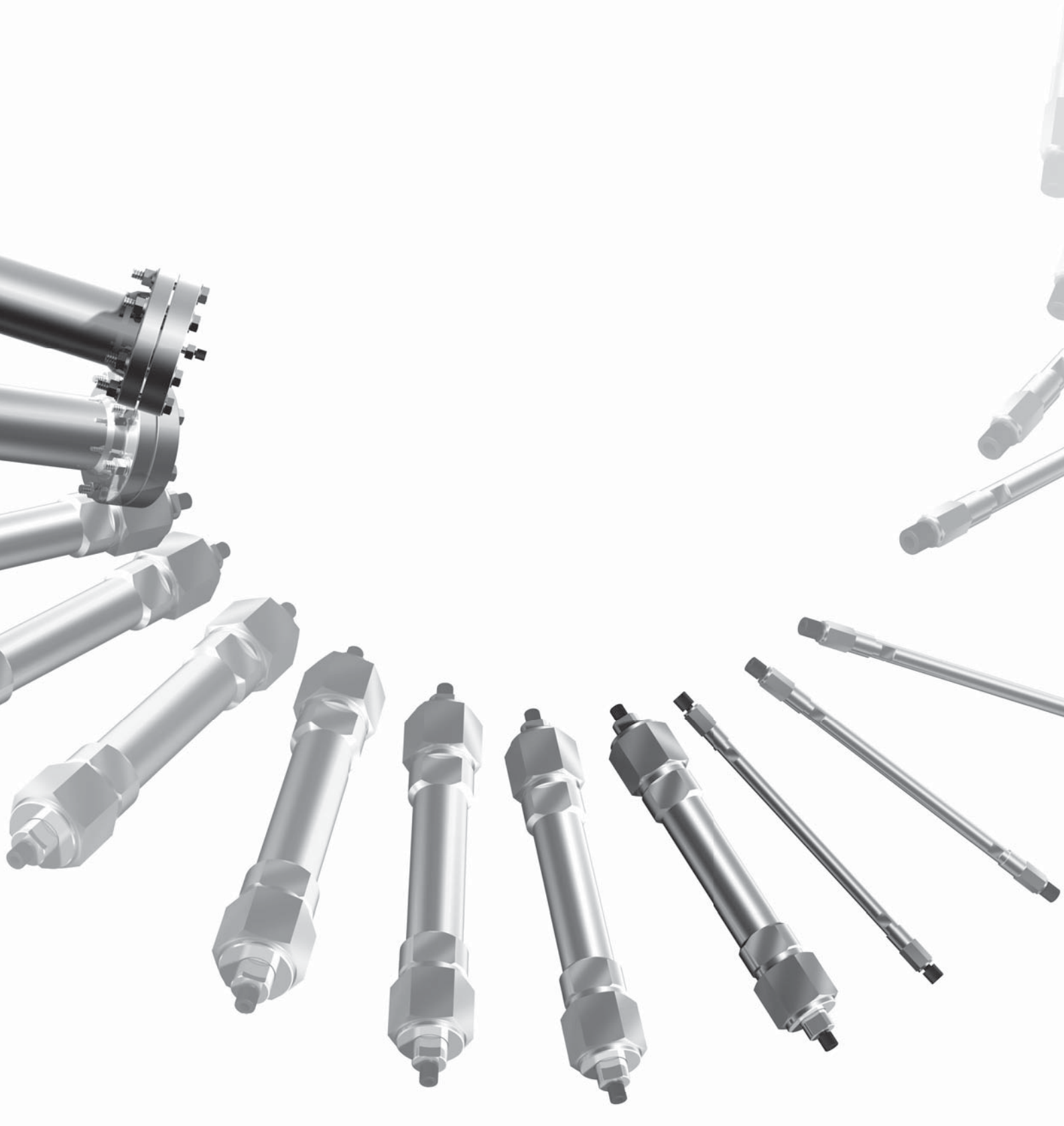
The selectivity for non–dissociative organic compounds on COSMOSIL 5C$_{18}$ AR–II and COSMOSIL 5C$_{18}$ AR is identical because the carbon content of both columns is the same.



COSMOSIL 5C$_{18}$–MS–II and COSMOSIL 5C$_{18}$–AR–II support the validation.

New Types of COSMOSIL columns are recommended for your new applications.

VII Index



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Packing Material's Name Index

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COSMOSIL C_{18}-MS-II

COSMOSIL C_{18}-MS-II

COSMOSIL C_{18}-MS-II

COSMOSIL C_{18}-MS-II

COSMOSIL C_{18}-AR-II

COSMOSIL C_{18}-AR-II

COSMOSIL C_{18}-AR-II

COSMOSIL C_{18}-PAQ

COSMOSIL C_{18}-PAQ

COSMOSIL Cholester

COSMOSIL Cholester

COSMOSIL πNAP

COSMOSIL PYE

COSMOSIL NPE

COSMOSIL PBB-R

COSMOSIL PBB-R

COSMOSIL CN-MS

COSMOSIL C_{22}-AR-II

COSMOSIL C_8-MS

COSMOSIL C_4-MS

COSMOSIL TMS-MS

COSMOSIL PE-MS

COSMOSIL SL-II

COSMOSIL HILIC

COSMOSIL HILIC

COSMOSIL Sugar-D

COSMOSIL NH_2-MS

COSMOSIL Protein-R

COSMOSIL Protein-R

COSMOSIL C_{18}-AR-300

COSMOSIL C_8-AR-300

COSMOSIL C_4-AR-300

COSMOSIL Ph-AR-300

COSMOSIL Diol-120-II

COSMOSIL Diol-300-II

COSMOGEL IEX Series

COSMOGEL IEX Series

COSMOSIL HIC

COSMOSIL Buckyprep

COSMOSIL Buckyprep-M

COSMOSIL PBB

COSMOSIL CNT-300

COSMOSIL CNT-1000

COSMOSIL CNT-1620

COSMOSIL C_{18}-OPN

COSMOSIL C_{18}-PREP

COSMOSIL SL-II -PREP

Silica gel 60

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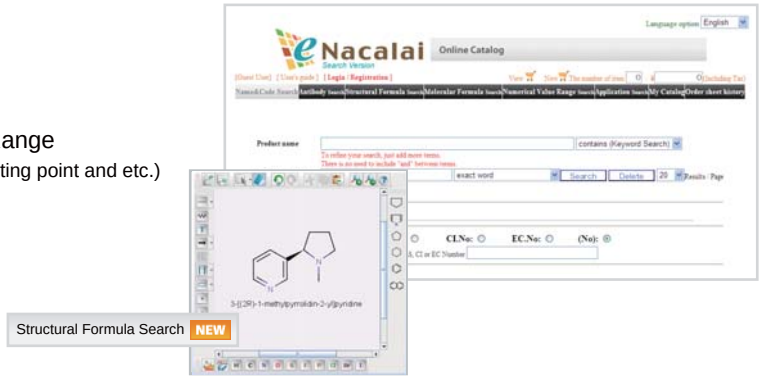

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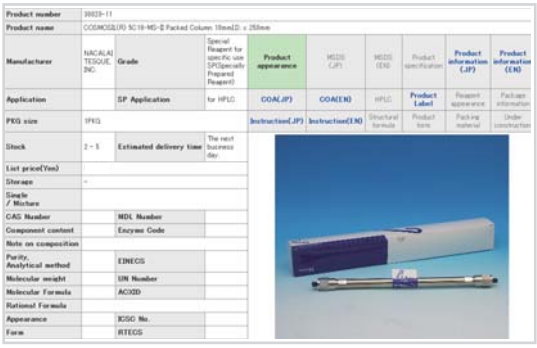

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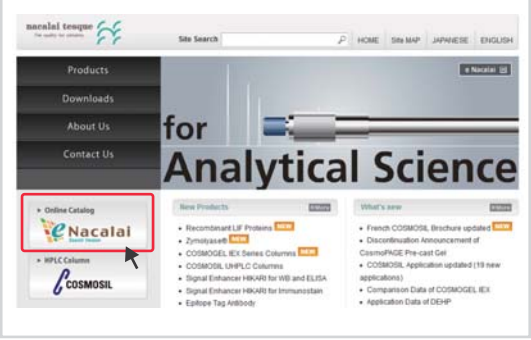

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NACALAI TESQUE, INC.

Nijo Karasuma, Nakagyo-ku, Kyoto 604-0855 JAPAN
TEL : +81 (0)75 251 1730
FAX : +81 (0)75 251 1763
E-mail : info.intl@nacalai.com

www.nacalai.com