

Rev. CO022020

Packed Columns for
Reversed Phase Chromatography
and Normal Phase Chromatography

TSK-GEL RPC Columns

TSK-GEL NPC Columns

INSTRUCTION MANUAL





TOSOH CORPORATION

Safety Precautions

To help protect you and/or your property from potential damage and ensure personal safety, please read this manual thoroughly before using the product.

[Notational Conventions]

| Notation | Explanation |
|--|--|
|  WARNING | Indicates a hazard with a medium level of risk which, if not avoided, could result in death or serious injury. |
|  CAUTION | Indicates a hazard with a low level of risk which, if not avoided, could result in minor or moderate injury. |

WARNING

■ **Keep away from fire**

Not taking proper precautions when using flammable solvents could result in fire, explosion, or poisoning.

CAUTION

■ **Use only in well-ventilated areas**

In case of insufficient ventilation, flammable and toxic solvents can cause fire, explosion, or poisoning.

■ **Do not spill solvents**

Spillage and leakage can cause fire, electric shock, poisoning, injury, and corrosion.

Wear appropriate protective gear when cleaning up a spill.

■ **Wear protective eye gear and gloves**

Organic solvents and acids should not come into direct contact with the skin.

■ **Handle the package with care**

Inappropriate handling may cause rupturing and/or splattering of the product.

■ **Only use this product for its intended use**

This product is intended for the separation and purification. Do not use it for any other purpose.

■ **Make sure compounds are safe**

Check that the target compounds and solutions after separation and purification are safe.

■ **Proper disposal**

Dispose in accordance with local laws and regulations.

NOTE

Keep this manual with the product for future reference.

Precautions: Shipping Solvents

| | | |
|----------------------|---------------------------------------|--|
| First Aid | Inhalation | <ul style="list-style-type: none"> • Move the person to an area with fresh air and rinse the mouth with plenty of water. • Call immediately for medical attention. |
| | Skin exposure | <ul style="list-style-type: none"> • Wash the exposed area with plenty of soap and water. |
| | Eye exposure | <ul style="list-style-type: none"> • Open the eyes as wide as possible and rinse with clean water for at least 15 minutes. • Call immediately for medical attention. |
| | Ingestion | <ul style="list-style-type: none"> • Rinse the mouth with plenty of water. • Call immediately for medical attention. |
| Handling and Storage | Ventilation | <ul style="list-style-type: none"> • Provide adequate air ventilation to keep organic vapor concentrations below approved level. |
| | Container handling | <ul style="list-style-type: none"> • Container may break if not handled with care. |
| | Wear appropriate protective equipment | <ul style="list-style-type: none"> • Use solvent-resistant gloves and protective eye gear when using this product. Use of gas mask, additional protective clothing or rubber boots could be appropriate when handling this product. |
| | Hazardous substance storage | <ul style="list-style-type: none"> • If any flammable solvents are used for shipping or storage of this product, keep away from fire or open heat sources. |
| Waste Disposal | Disposal methods | <ul style="list-style-type: none"> • Dispose in accordance with local laws and regulations. |
| | General considerations | <ul style="list-style-type: none"> • Please pay attention to all safety precautions with respect to the handling and storage of this product. |
| | Disposal precautions | <ul style="list-style-type: none"> • Assure that appropriate countermeasures are taken when incinerating solvents that contain acetonitrile. Fumes produced during incineration may contain nitrogen oxides. |

Shipping solvent for each column : See Section 1. General Information.

Precautions: Packing Material

| | | |
|----------------------|---------------------------------------|--|
| First Aid | Inhalation | <ul style="list-style-type: none"> • Move the person to an area with fresh air and rinse the mouth with plenty of water. • Call immediately for medical attention. |
| | Skin exposure | <ul style="list-style-type: none"> • Wash the exposed area with plenty of soap and water. |
| | Eye exposure | <ul style="list-style-type: none"> • Open the eyes as wide as possible and rinse with clean water for at least 15 minutes. • Call immediately for medical attention. |
| | Ingestion | <ul style="list-style-type: none"> • Rinse the mouth with plenty of water. • Call immediately for medical attention. |
| Handling and Storage | Ventilation | <ul style="list-style-type: none"> • Provide adequate air ventilation to keep organic vapor concentrations below approved level. |
| | Container handling | <ul style="list-style-type: none"> • Container may break if not handled with care. |
| | Wear appropriate protective equipment | <ul style="list-style-type: none"> • Use solvent-resistant gloves and protective eye gear when using this product. Use of gas mask, additional protective clothing or rubber boots could be appropriate when handling this product. |
| | Hazardous substance storage | <ul style="list-style-type: none"> • If any flammable solvents are used for shipping or storage of this product, keep away from fire or open heat sources. |
| Waste Disposal | Fire precautions | <ul style="list-style-type: none"> • Do not expose the chromatographic resin to fire or open heat sources. |
| | Disposal methods | <ul style="list-style-type: none"> • Dispose in accordance with local laws and regulations. |
| | General considerations | <ul style="list-style-type: none"> • Please pay attention to all safety precautions with respect to the handling and storage of this product. |
| | Disposal precautions | <ul style="list-style-type: none"> • Appropriate nitrogen oxides exhaust emission precautions should be taken specifically for TSKgel CN-80T_s and TSKgel NH₂-60. |

- Polymer gel
TSKgel Octadecyl-2PW, TSKgel Octadecyl-4PW, TSKgel Octadecyl-NPR, TSKgel Phenyl-5PW RP, TSKgel Phenyl-5PW RP Glass, TSKgel Enviropak G1, TSKgel VMPak-25
- Modified silica gel
TSKgel ODS-100S, TSKgel ODS-140HTP 2.3 μ m, TSKgel Super-ODS, TSKgel ODS-80T_M, TSKgel ODS-80T_s, TSKgel ODS-120A, TSKgel ODS-120T, TSKgel CN-80T_s, TSKgel Super-Octyl, TSKgel Octyl-80T_s, TSKgel Super-Phenyl, TSKgel NH₂-60, TSKgel OH-120, TSKgel TMS-250, TSKgel OligoDNA RP
- Silica gel
TSKgel Silica-60, TSKgel Silica-150

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1. General Information

TSK-GEL RPC/NPC columns have been optimized for high performance. Typical application fields of TSK-GEL RPC/NPC columns are listed in Table 1. Please read this INSTRUCTION MANUAL carefully and use the column as recommended in order to make effective use of its high performance.

Table 1 Descriptions of TSK-GEL RPC/NPC Columns

| Type | Description | Shipping Solvent | Application |
|-------------------------------|--|--|--|
| TSKgel ODS-120A | ODS bonded silica gel | CH ₃ OH/H ₂ O=7/3 | Drugs in blood, Detergents Polyphenols |
| TSKgel ODS-120T | Fully capped ODS bonded silica gel | | Peptides, Low molecular proteins |
| TSKgel Super-ODS | | | Low molecular bioorganisms |
| TSKgel ODS-80T _M | | | |
| TSKgel ODS-80T _S | | | |
| TSKgel ODS-100S | | | |
| TSKgel ODS-140HTP 2.3μm | | CH ₃ CN | |
| TSKgel Super-Octyl | Fully capped, Octyl bonded silica gel | CH ₃ OH/H ₂ O=7/3 | Peptides, Nucleotides Amino acids, Pharmaceuticals |
| TSKgel Octyl-80T _S | | | |
| TSKgel Super-Phenyl | Fully capped, Phenyl bonded silica gel | CH ₃ CN/H ₂ O=5/5 | Peptides, Pharmaceuticals |
| TSKgel CN-80T _S | Fully capped, CN bonded silica gel | CH ₃ CN/H ₂ O=4/6 | |
| TSKgel Silica-60 | Spherical silica gel | n-C ₈ H ₁₄ /C ₂ H ₅ OH=8/2 | Various isomer, Drugs |
| TSKgel Silica-150 | | | Polyphenol, Lipid Vitamines, Basic substance |
| TSKgel NH ₂ -60 | NH ₂ bonded silica gel | CH ₃ OH | Monosaccharides, Disaccharides, Oligosaccharides |
| TSKgel OH-120 | OH bonded silica gel | n-C ₈ H ₁₄ /C ₂ H ₅ OH=8/2 | Steroides |
| TSKgel TMS-250 | Fully capped, C ₁ bonded silica gel | CH ₃ OH/H ₂ O=5/5 | High molecular proteins |

Table 1 Descriptions of TSK-GEL RPC/NPC Columns

(continued)

| Type | Description | Shipping Solvent | Application |
|--|---|--|-----------------------------|
| TSKgel OligoDNA RP | Fully capped, ODS bonded silica gel | CH ₃ CN/H ₂ O=5/5 | OligoDNA |
| TSKgel Octadecyl-NPR | Octadecyl, Phenyl groups bonded hydrophilic polymer gel | CH ₃ OH/H ₂ O=7/3 | Peptides, Proteins, Enzymes |
| TSKgel Octadecyl-2PW | | CH ₃ CN/H ₂ O=55/45 | Drugs |
| TSKgel Octadecyl-4PW | | CH ₃ OH/H ₂ O=6/4 | Peptides |
| TSKgel Enviropak G1 | | CH ₃ CN (50 mmol/L) /KH ₂ PO ₄ =5/5 | Agricultural chemicals |
| TSKgel Phenyl-5PW RP TSKgel Phenyl-5PW RP Glass | | CH ₃ OH/H ₂ O=3/7 | Proteins, Enzymes |
| TSKgel VMPak-25 | Hydrophilic polymer gel | H ₂ O | Drugs |

2. Unpacking

Check that there is no visible damage to the outer package or the column.



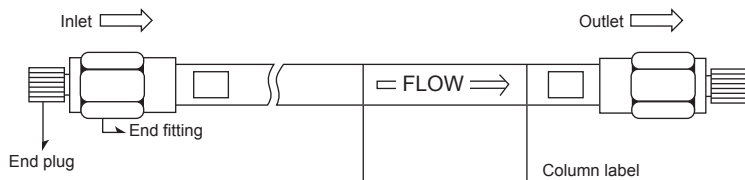
Figure 1 Appearance of the Package

Check that the following documents are shipped with the column.

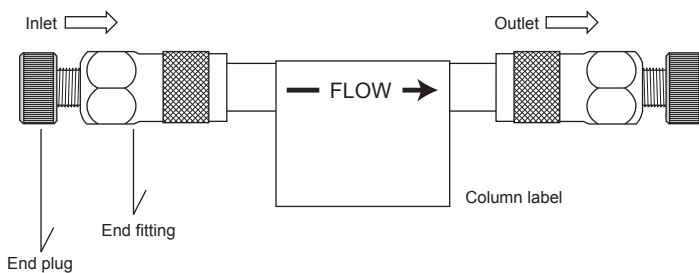
- 1) INSTRUCTION MANUAL 1 copy
- 2) INSPECTION DATA 1 copy

3. Column Parts

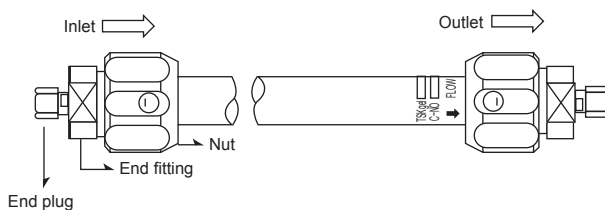
* For TSKgel Super-ODS, TSKgel Super-Octyl, TSKgel Super-Phenyl and Semi-micro column



* For TSKgel ODS-140HTP 2.3 μ m



* For TSKgel Phenyl-5PW RP Glass (Glass column)



* For the other columns

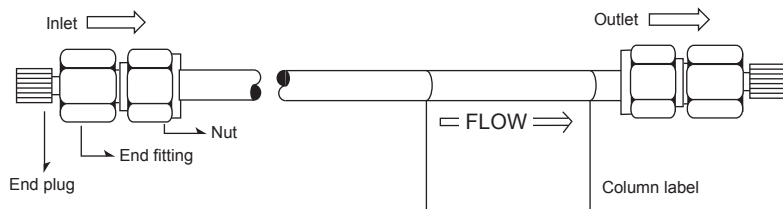


Figure 2 Column Parts

4. Column Installation

- (1) Confirm the correct product name is listed on the column label.
- (2) Each column is equipped with a union nut enabling a connection to a 1/16"O.D. capillary tubing. The union nut is designed for American standard compression plugs and ferrules.
- (3) Confirm the flow direction on the column label or etched onto the column as shown in Figure 2. Solvent should flow only into the column from the inlet side. The columns are designed so that optimal resolution is obtained when the flow direction is as indicated on the column.
- (4) Purge all air out of the tubing using the mobile phase. This helps to prevent any air from entering the column. Any air in the tubing causes serious deterioration of column efficiency.
- (5) Initially set the solvent flow rate at one-half of the intended flow rate. Make sure that the solvent is flowing freely out of the end of the tubing from the injector.
- (6) Remove the end plugs from the column and connect the inlet of the column to the tubing from the injector. Make sure that the tubing is fully inserted into the compression fittings before tightening in order to minimize dead volume. Always keep dead volume to the absolute minimum throughout the entire system.
- (7) After the solvent flows from the outlet of the column, connect the column to the detector.
- (8) Start pumping the solvent at a flow rate less than one-half of the final flow rate. Avoid a sudden pressure surge to the column.
- (9) The columns are very sensitive to pressure pulsing. A pulseless pumping system should be used.
- (10) The columns should be equilibrated before use by allowing at least 10 column volumes of solvent to pass through the column.

5. Column Maintenance

- (1) If the column is used in routine daily operation, it is permissible to leave the mobile phase in the column overnight if the mobile phase is not corrosive. If halides are included in the mobile phase, it is better to replace the mobile phase with a suitable solvent (for example "Packed Solvent" shown in the INSPECTION DATA sheet) even for one night.
- (2) If the column will not be used for several days, it should be stored as follows:
 - a) Purge the system with the "Packed Solvent" shown in the INSPECTION DATA sheet at a flow rate one-half of the operating flow rate as shown in Table 2. (Purge the system with distilled or ion-exchanged water if you have used a buffer solution, as a mobile phase, which contains salt in considerably high concentrations.)
 - b) Remove the column from the system and keep the ends of the column tightly capped with the end plugs supplied with the column.

- c) Store the column at a relatively constant temperature in its original shipping container. Take care not to allow the column to freeze during storage.
- (3) The performance of the guard column may be decreased by repeated removal from the cartridge holder. TOSOH recommends that the guard column is kept in the cartridge holder with both ends capped with end plugs.

6. Solvent Selection and Preparation

- (1) The shipping solvent of each individual column is shown in Table 1.
Before using the column, the solvent should be replaced with an appropriate mobile phase for analysis. Solvent replacement should be performed at a flow rate one-half of the normal operating flow rate, or at a pressure below the maximum pressure shown in Table 2. Note that a drastic change of solvent composition or frequent solvent replacements may shorten the lifetime of the column.
- (2) Keep the pH range of the solvent within the following range:
- pH 2.0 ~ 7.5 ... TSKgel Super-ODS, TSKgel Super-Octyl, TSKgel Super-Phenyl, TSKgel ODS-80T_M, TSKgel ODS-80T_S, TSKgel ODS-120A, TSKgel ODS-120T, TSKgel ODS-100S, TSKgel ODS-140HTP 2.3 μ m, TSKgel Octyl-80T_S, TSKgel CN-80T_S, TSKgel Silica-60, TSKgel Silica-150, TSKgel NH₂-60, TSKgel OH-120, TSKgel TMS-250 and TSKgel OligoDNA RP
- pH 2.0 ~ 12.0 ... TSKgel Octadecyl-NPR, TSKgel Octadecyl-2PW, TSKgel Octadecyl-4PW, TSKgel Enviropak G1 and TSKgel Phenyl-5PW RP

The pH range should be selected based on the stability of both packing material and the column itself. At a pH below pH 2.0, the ligand binding sites on the silica-based support are subject to hydrolysis by acidic solutions. Above pH 7.5, the silica backbone may dissolve, leading to rapid column failure. Additionally, the stainless steel of the column is subject to corrosion at a low pH particularly when using halides.

- (3) The solvent should be filtered through a 0.5 μ m filter in order to prevent the accumulation of small particles.
The performances of semi-micro columns quickly deteriorates when exposed to small-particle contamination. Thus it is highly recommended that an in-line filter containing a membrane of 0.2 μ m ~ 0.5 μ m pore size is inserted between the pump and the sample injector.

Line filter

Part No. 0014594 Filter assembly

Part No. 0006280 Fluoropore filter (0.45 μ m, package of 100)

- (4) Solvents should be degassed to ensure optimal flow through the system.

7. Flow Rate

The flow rate should be selected based on the desired resolution, column life and assay time. Although the TSK-GEL RPC/NPC columns are designed for high-speed analysis, TOSOH recommends that these columns are operated at a rather low flow rate because better resolution and extended column life can be expected. A suitable flow rate and the maximum flow rate depend on the organic solvent in the mobile phase. When using the column for the first time, the flow rate should be set at a linear velocity of 6 cm/min (0.05 mL/min for 1.0 mm(I.D.), 0.20 mL/min for 2.0 mm(I.D.), 0.43 mL/min for 3.0 mm(I.D.) and 1.00 mL/min for 4.6 mm(I.D.)).

The column life may be reduced if the column is operated near the maximum pressure.

The maximum pressures are shown in Table 2. The viscosity of the solvent must be considered when selecting the flow rate, too.

Table 2 Maximum Pressure

| Part No. | Type | Column Size mm(I.D.)×cm(L) | Maximum Pressure (MPa) |
|----------|-------------------------------|-------------------------------|------------------------------|
| 0021927 | TSKgel ODS-140HTP 2.3 μ m | 2.1×5 | 60.0 |
| 0021928 | / | 2.1×10 | 60.0 |
| 0019538 | TSKgel ODS-100S | 2.0×15 | 15.0 |
| 0019539 | / | 2.0×25 | 20.0 |
| 0019536 | / | 3.0×15 | 15.0 |
| 0019537 | / | 3.0×25 | 20.0 |
| 0019534 | / | 4.6×15 | 15.0 |
| 0019535 | / | 4.6×25 | 20.0 |
| 0020015 | TSKgel Super-ODS | 1.0×5 | 15.0 |
| 0019541 | / | 2.0×5 | 25.0 |
| 0019542 | / | 2.0×10 | 25.0 |
| 0018154 | / | 4.6×5 | 30.0 |
| 0018197 | / | 4.6×10 | 30.0 |
| 0016651 | TSKgel ODS-80T _M | 4.6×7.5 | 10.0 |
| 0019017 | / | 4.6×10 | 15.0 |
| 0008148 | / | 4.6×15 | 20.0 |
| 0008149 | / | 4.6×25 | 30.0 |
| 0014000 | / | 6.0×15 | 15.0 |
| 0014001 | / | 7.8×30 | 7.5 |
| 0014002 | / | 21.5×30 | 6.0 |
| 0018150 | TSKgel ODS-80T _S | 2.0×15 | 20.0 |
| 0018151 | / | 2.0×25 | 30.0 |
| 0017200 | / | 4.6×7.5 | 10.0 |
| 0017201 | / | 4.6×15 | 20.0 |
| 0017202 | / | 4.6×25 | 30.0 |
| 0017203 | / | 6.0×15 | 15.0 |
| 0018409 | / | 20.0×25 | 15.0 |
| 0017380 | / | 21.5×30 | 6.0 |
| 0007636 | TSKgel ODS-120A | 4.6×15 | 15.0 |
| 0007124 | / | 4.6×25 | 20.0 |
| 0014003 | / | 6.0×15 | 10.0 |
| 0007129 | / | 7.8×30 | 7.5 |
| 0006172 | / | 21.5×30 | 6.0 |

Table 2 Maximum Pressure

(continued)

| Part No. | Type | Column Size mm(I.D.)×cm(L) | Maximum Pressure (MPa) |
|----------|-----------------------------|-------------------------------|------------------------------|
| 0018152 | TSKgel ODS-120T | 2.0×15 | 15.0 |
| 0018153 | ∕ | 2.0×25 | 20.0 |
| 0007637 | ∕ | 4.6×15 | 15.0 |
| 0007125 | ∕ | 4.6×25 | 20.0 |
| 0014004 | ∕ | 6.0×15 | 10.0 |
| 0007130 | ∕ | 7.8×30 | 7.5 |
| 0007134 | ∕ | 21.5×30 | 6.0 |
| 0020013 | TSKgel Super-Octyl | 2.0×5 | 15.0 |
| 0020014 | ∕ | 2.0×10 | 30.0 |
| 0018275 | ∕ | 4.6×5 | 30.0 |
| 0018276 | ∕ | 4.6×10 | 30.0 |
| 0017344 | TSKgel Octyl-80Ts | 4.6×15 | 20.0 |
| 0017345 | ∕ | 4.6×25 | 30.0 |
| 0017346 | ∕ | 6.0×15 | 15.0 |
| 0017348 | TSKgel CN-80Ts | 4.6×15 | 20.0 |
| 0017349 | ∕ | 4.6×25 | 30.0 |
| 0017350 | ∕ | 6.0×15 | 15.0 |
| 0020017 | TSKgel Super-Phenyl | 2.0×5 | 8.0 |
| 0020018 | ∕ | 2.0×10 | 15.0 |
| 0018277 | ∕ | 4.6×5 | 30.0 |
| 0018278 | ∕ | 4.6×10 | 30.0 |
| 0007147 | TSKgel Silica-60 | 4.6×25 | 15.0 |
| 0006730 | ∕ | 21.5×30 | 2.5 |
| 0007146 | TSKgel Silica-150 | 4.6×25 | 15.0 |
| 0006723 | ∕ | 21.5×30 | 2.5 |
| 0007127 | TSKgel NH ₂ -60 | 4.6×25 | 20.0 |
| 0007128 | TSKgel OH-120 | 4.6×25 | 20.0 |
| 0007190 | TSKgel TMS-250 | 4.6×7.5 | 2.0 |
| 0013352 | TSKgel OligoDNA RP | 4.6×15 | 12.0 |
| 0013353 | ∕ | 7.8×15 | 12.0 |
| 0014005 | TSKgel Octadecyl-NPR* | 4.6×3.5 | 20.0 |
| 0018754 | TSKgel Octadecyl-2PW* | 2.0×15 | 7.0 |
| 0017500 | ∕ | 4.6×15 | 10.0 |
| 0017501 | ∕ | 6.0×15 | 10.0 |
| 0018755 | TSKgel Octadecyl-4PW* | 2.0×15 | 10.0 |
| 0013351 | ∕ | 4.6×15 | 12.0 |
| 0016257 | ∕ | 21.5×15 | 2.5 |
| 0017794 | TSKgel Enviropak G1* | 6.0×15 | 10.0 |
| 0020011 | TSKgel VMpak-25* | 2.0×5 | 2.0 |
| 0020012 | ∕ | 2.0×15 | 6.0 |
| 0018756 | TSKgel Phenyl-5PW RP* | 2.0×7.5 | 1.0 |
| 0008043 | ∕ | 4.6×7.5 | 3.0 |
| 0016260 | ∕ | 21.5×15 | 3.0 |
| 0014006 | TSKgel Phenyl-5PW RP Glass* | 5.0×5 | 2.0 |
| 0014007 | ∕ | 8.0×7.5 | 2.0 |

Note : The columns with the symbol "*" are of polymer-based gel and the others are of silica-based gel.

8. Temperature

The optimal operating temperature for each column is as follows.

- 4 °C ~ 30 °C TSKgel Phenyl-5PW RP Glass
- 4 °C ~ 60 °C TSKgel Phenyl-5PW RP and TSKgel Octadecyl-NPR
- 5 °C ~ 50 °C TSKgel Octadecyl-2PW and TSKgel Enviropak G1
- 10 °C ~ 50 °C TSKgel Super-ODS, TSKgel Super-Octyl, TSKgel Super-Phenyl,
TSKgel ODS-80T_M, TSKgel ODS-80T_S, TSKgel ODS-120A,
TSKgel ODS-120T, TSKgel ODS-100S,
TSKgel ODS-140HTP 2.3 μm, TSKgel Octyl-80T_S,
TSKgel CN-80T_S, TSKgel Silica-60, TSKgel Silica-150,
TSKgel NH₂-60, TSKgel OH-120, TSKgel TMS-250,
TSKgel OligoDNA RP and TSKgel Octadecyl-4PW

9. Sample Preparation

(1) Preparation of Sample Solution

Prepare a sample solution immediately prior to injection by dissolving the sample into the solvent that is used as an eluent. The eluent should be optimized by adjusting the pH, salt concentration, etc. so that the sample is completely dissolved, otherwise the column lifetime may be reduced by unexpected precipitation of sample on the column.

(2) Filtration of Insoluble Particles

The sample solution should be filtered with a micropore-filter (0.5 μm). Even though no particles can be detected by the naked eye, insoluble particles may exist in the sample.

10. Measurement of Number of Theoretical Plates and Asymmetry Factor

(1) Number of theoretical plates (N)

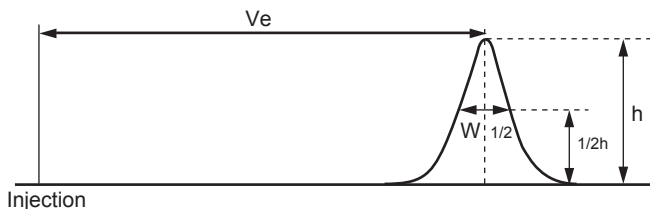


Figure 3 Calculation of Number of Theoretical Plates

The N is calculated using an unretained molecule by the half-peak width method as shown in Figure 3 and the following equation.

$$N=5.54(Ve/W_{1/2})^2$$

where:

Ve : Elution time

W_{1/2} : Width of peak at half-height

h : Peak height

N : Number of theoretical plates/column

(2) Asymmetry factor (As)

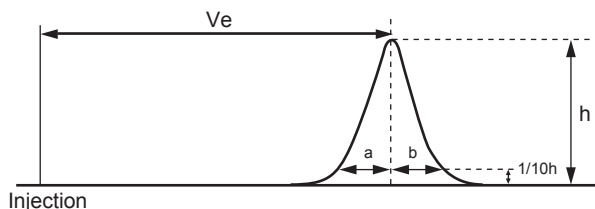


Figure 4 Calculation of Asymmetry Factor

The asymmetry factor is calculated according to Figure 4 and the following equation.

$$As=b/a$$

- (3) The N and As should be measured with an instrument with small dead volume.
- (4) The N and As are mentioned in the INSPECTION DATA sheet together with the experimental conditions.

11. Guard Column

Fundamental keys to prevent problems have been outlined in Section 4 to 9. When impurities that tend to adsorb onto the packing material are present in a sample, they are typically adsorbed at the inlet side of the column and gradually accumulate causing a reduction in the number of theoretical plates and a decrease in column performance.

In such cases the original column performance can be maintained by connecting a guard column between the injection valve and the analytical column. The guard column should be replaced when the performance deteriorates as a result of the adsorption of such a material to the guard column. A guard column can not be used in place of analytical column.

The use of a guard column will not improve the resolution obtained on the analytical column.

11-1 Effect of Guard Column Installation

- (1) Contamination of the analytical column can be prevented by the removal of adsorptive or insoluble materials in the sample.
- (2) Pressure shock, due to pump pulsation, to the analytical column should be avoided.

11-2 Type and Selection of Guard Columns

Guard columns specifications are shown in Table 3 to 6.

Table 3 Guard Column

| Part No. | Type | Column Size mm(I.D.)×cm(L) | Applied Column mm(I.D.)×cm(L) |
|----------|-------------------------------------|-------------------------------|--|
| 0014098 | TSKguardcolumn ODS-80T _M | 21.5×7.5 | TSKgel ODS-80T _M (21.5×30) |
| 0007642 | TSKguardcolumn ODS | 21.5×7.5 | TSKgel ODS-120A (21.5×30) TSKgel ODS-120T (21.5×30) |
| 0017385 | TSKguardcolumn ODS-80T _S | 21.5×7.5 | TSKgel ODS-80T _S (21.5×30) |
| 0018410 | ∕ | 20.0×5 | TSKgel ODS-80T _S (20.0×25) |
| 0017383 | ∕ | 45.0×5 | TSKgel ODS-80T _S (55.0×30, 55.0×60) |
| 0018207 | G-filter for guard holder | 4.0×0.4 | TSKgel Super-ODS |
| 0017795 | TSKguardcolumn Enviropak G1 | 6.0×1 | TSKgel Enviropak G1 |
| 0017502 | TSKguardcolumn Octadecyl-2PW | 4.6×1 | TSKgel Octadecyl-2PW (4.6×15) |
| 0017503 | ∕ | 6.0×1 | TSKgel Octadecyl-2PW (6.0×15) |

Table 4 Cartridge Column

| Part No. | Type | Column Size mm(I.D.)×cm(L) | Applied Column mm(I.D.)×cm(L) |
|----------|------------------------------------|-------------------------------|---|
| 0019004 | TSKguardgel ODS-80T _M | 3.2×1.5 | TSKgel ODS-80T _M (4.6×15, 4.6×25, 6.0×15) |
| 0019011 | TSKguardgel ODS-80T _S | 3.2×1.5 | TSKgel ODS-80T _S (4.6×15, 4.6×25, 6.0×15) |
| 0019005 | TSKguardgel ODS-120A | 3.2×1.5 | TSKgel ODS-120A (4.6×15, 4.6×25, 6.0×15) |
| 0019006 | TSKguardgel ODS-120T | 3.2×1.5 | TSKgel ODS-120T (4.6×15, 4.6×25, 6.0×15) |
| 0019012 | TSKguardgel Octyl-80T _S | 3.2×1.5 | TSKgel Octyl-80T _S (4.6×15, 4.6×25, 6.0×15) |
| 0019013 | TSKguardgel CN-80T _S | 3.2×1.5 | TSKgel CN-80T _S (4.6×15, 4.6×25, 6.0×15) |
| 0019007 | TSKguardgel Phenyl-5PW RP | 3.2×1.5 | TSKgel Phenyl-5PW (4.6×7.5) |
| 0019008 | TSKguardgel Octadecyl-4PW | 3.2×1.5 | TSKgel Octadecyl-4PW (4.6×15) |
| 0019325 | TSKguardgel ODS-S1 | 2.0×1 | Semi-micro column for ODS type (2.0×15, 2.0×25) |
| 0019540 | TSKguardgel ODS-100S | 3.2×1.5 | TSKgel ODS-100S (3.0×15, 3.0×25, 4.6×15, 4.6×25) |
| 0019672 | TSKguardgel Super-ODS | 2.0×1 | TSKgel Super-ODS (2.0×5, 2.0×10) |

Note : Three cartridge columns are packed in a box.

Table 5 Cartridge Holder

| Part No. | Type | Column Size mm(I.D.)×cm(L) | Applied Column mm(I.D.)×cm(L) |
|----------|-----------------------------------|-------------------------------|------------------------------------|
| 0019018 | Cartridge Holder (for 3.2×1.5) | 3.2×1.5 | Cartridge Column |
| 0018206 | Guard Holder | 4.0×0.4 | G-filter (for TSKgel Super-ODS) |
| 0019308 | Cartridge Holder (for 2.0×1) | 2.0×1 | Cartridge Column |

Note : Two nuts, two ferrules and two small pieces of tubing are attached to the cartridge holder as accessories.

In addition, column-extracting tool is attached to Part No. 0019018.

Table 6 Guardgel Kit (Glass Column)

| Part No. | Type | Column Size mm(I.D.)×cm(L) | Applied Column mm(I.D.)×cm(L) |
|----------|--|-------------------------------|---|
| 0014022 | TSKguardgel Phenyl-5PW RP Kit Glass | 8.0×1 | TSKgel Phenyl-5PW RP Glass (8.0×7.5) |

11-3 Guard Column Replacement

Since the guard column has limited adsorption capacity, it has a finite lifetime.

The guard column must be replaced before contamination extends to the main analytical column.

The frequency of the guard column replacement can not be standardized because it depends on various factors such as application (analysis or preparative separation), sample properties (properties of principal components, properties and concentrations of impurities, etc.), sample loading, solvents, flow rate, etc.

Since an increase in the system pressure during operation could indicate clogging at the end fitting of the guard column or contamination of the gel, it is a good idea to replace the guard column when the pressure has increased.

In general, when changes in the results are observed, the guard column should be replaced immediately.

11-4 Bulk Gel

The list of the bulk gels are shown in Table 7.

Table 7 Bulk Gel

| Type | Particle Size 5 μ m | | | | Particle Size 10 μ m | |
|-------------------------------|-------------------------|--------|----------|--------|--------------------------|--------|
| | Part No. | Weight | Part No. | Weight | Part No. | Weight |
| TSKgel ODS-80T _M | 0008173 | 5 g | 0008174 | 10 g | 0014091 | 10 g |
| TSKgel ODS-80T _s | 0017319 | 5 g | 0017318 | 10 g | 0017320 | 10 g |
| TSKgel ODS-120A | 0007115 | 5 g | 0006995 | 10 g | 0006996 | 10 g |
| TSKgel ODS-120T | 0007116 | 5 g | 0007119 | 10 g | 0007121 | 10 g |
| TSKgel Octyl-80T _s | 0017372 | 5 g | 0017373 | 10 g | 0017374 | 10 g |
| TSKgel CN-80T _s | 0017375 | 5 g | 0017376 | 10 g | 0017377 | 10 g |
| TSKgel TMS-250 | — | — | — | — | 0007122 | 5 g |
| TSKgel NH ₂ -60 | 0007117 | 5 g | 0007120 | 10 g | 0007123 | 10 g |
| TSKgel OH-120 | 0007118 | 5 g | 0005578 | 10 g | 0005784 | 10 g |
| TSKgel Silica-150 | 0007143 | 5 g | 0005189 | 10 g | 0005190 | 10 g |
| TSKgel Silica-60 | 0007144 | 5 g | 0006219 | 10 g | 0007145 | 10 g |

12. Troubleshooting

(1) Clogging of the inlet filter

Increased pressure or decreased flow rate are indicative of a clogged inlet filter. In this case, clean the end fitting by reversing the flow direction through the column. (The flow rate must be kept below one-half of the operating flow rate.)

(2) Contamination

Continuous column operation may lead to gradual accumulation of strongly ionic compounds or hydrophobic compounds.

This is demonstrated by changes in chromatographic behavior and loss of resolution. Adsorbed materials may be removed from the column by injections of solvent with different properties from the operating mobile phase.

Recommended column cleaning solutions are shown in Section 14.

(3) Bed Compression

Failure to properly clean the analytical column may result in the formation of a void at the column head due to bed compression.

This failure can be confirmed by carefully removing the column end and inspecting the bed. Any void should be filled with slurry of the appropriate RPC/NPC gel.

13. Quality Specification and Warranty

13-1 INSPECTION DATA

The inspection conditions and the results of each individual column are shown on the INSPECTION DATA sheet. The number of theoretical plates is expressed as the number per column.

The inspection results are different for each column.

13-2 Quality Specifications

TSK-GEL RPC/NPC columns are delivered according to the specifications as shown in Table 8.

13-3 Warranty

Upon receiving the column, check that the column is not damaged and test the performance according to Section 10. If the guaranteed specifications in Table 8 can not be obtained, contact a local TOSOH representative within two weeks. Note that the column lifetime is not guaranteed.

Table 8 Guaranteed Specifications

| Part No. | Type | Column Size mm(I.D.)×cm(L) | Number of Theoretical Plates (TP/Column) | Asymmetry Factor |
|----------|-------------------------------|-------------------------------|--|---------------------|
| 0019538 | TSKgel ODS-100S | 2.0×15 | 10,000 | 0.90~1.25 |
| 0019539 | 〃 | 2.0×25 | 16,500 | 0.90~1.25 |
| 0019536 | 〃 | 3.0×15 | 11,000 | 0.90~1.25 |
| 0019537 | 〃 | 3.0×25 | 18,000 | 0.90~1.25 |
| 0019534 | 〃 | 4.6×15 | 11,000 | 0.90~1.25 |
| 0019535 | 〃 | 4.6×25 | 18,000 | 0.90~1.25 |
| 0021927 | TSKgel ODS-140HTP 2.3 μ m | 2.1×5 | 7,000 | 0.90~1.30 |
| 0021928 | 〃 | 2.1×10 | 14,000 | 0.90~1.30 |
| 0020015 | TSKgel Super-ODS | 1.0×5 | 1,500 | 0.8~2.0 |
| 0019541 | 〃 | 2.0×5 | 6,000 | 0.8~1.5 |
| 0019542 | 〃 | 2.0×10 | 12,000 | 0.8~1.5 |
| 0018154 | 〃 | 4.6×5 | 8,000 | 0.8~1.6 |
| 0018197 | 〃 | 4.6×10 | 16,000 | 0.8~1.6 |
| 0016651 | TSKgel ODS-80T _M | 4.6×7.5 | 4,500 | 0.7~1.8 |
| 0019017 | 〃 | 4.6×10 | 6,000 | 0.7~1.8 |
| 0008148 | 〃 | 4.6×15 | 11,000 | 0.7~1.8 |
| 0008149 | 〃 | 4.6×25 | 18,000 | 0.7~1.8 |
| 0014000 | 〃 | 6.0×15 | 11,000 | 0.7~1.8 |
| 0014001 | 〃 | 7.8×30 | 6,000 | 0.7~1.9 |
| 0014002 | 〃 | 21.5×30 | 6,000 | 0.7~1.9 |
| 0018150 | TSKgel ODS-80T _S | 2.0×15 | 11,000 | 0.8~1.6 |
| 0018151 | 〃 | 2.0×25 | 18,000 | 0.8~1.6 |
| 0017200 | 〃 | 4.6×7.5 | 4,500 | 0.7~1.8 |
| 0017201 | 〃 | 4.6×15 | 11,000 | 0.7~1.8 |
| 0017202 | 〃 | 4.6×25 | 18,000 | 0.7~1.8 |
| 0017203 | 〃 | 6.0×15 | 11,000 | 0.7~1.8 |
| 0018409 | 〃 | 20.0×25 | 22,000 | 0.7~1.8 |
| 0017380 | 〃 | 21.5×30 | 6,000 | 0.7~1.8 |
| 0007636 | TSKgel ODS-120A | 4.6×15 | 7,000 | 0.7~1.8 |
| 0007124 | 〃 | 4.6×25 | 10,000 | 0.7~1.8 |
| 0014003 | 〃 | 6.0×15 | 7,000 | 0.7~1.8 |
| 0007129 | 〃 | 7.8×30 | 6,000 | 0.7~1.9 |
| 0006172 | 〃 | 21.5×30 | 6,000 | 0.7~1.9 |
| 0018152 | TSKgel ODS-120T | 2.0×15 | 6,500 | 0.8~1.7 |
| 0018153 | 〃 | 2.0×25 | 10,000 | 0.8~1.7 |
| 0007637 | 〃 | 4.6×15 | 7,000 | 0.7~1.8 |
| 0007125 | 〃 | 4.6×25 | 10,000 | 0.7~1.8 |
| 0014004 | 〃 | 6.0×15 | 7,000 | 0.7~1.8 |
| 0007130 | 〃 | 7.8×30 | 6,000 | 0.7~1.9 |
| 0007134 | 〃 | 21.5×30 | 6,000 | 0.7~1.9 |
| 0020013 | TSKgel Super-Octyl | 2.0×5 | 1,500 | 0.7~2.0 |
| 0020014 | 〃 | 2.0×10 | 5,000 | 0.8~1.8 |
| 0018275 | 〃 | 4.6×5 | 8,000 | 0.8~1.6 |
| 0018276 | 〃 | 4.6×10 | 16,000 | 0.8~1.6 |
| 0017344 | TSKgel Octyl-80T _S | 4.6×15 | 11,000 | 0.7~1.8 |
| 0017345 | 〃 | 4.6×25 | 18,000 | 0.7~1.8 |
| 0017346 | 〃 | 6.0×15 | 11,000 | 0.7~1.8 |
| 0017348 | TSKgel CN-80T _S | 4.6×15 | 11,000 | 0.7~1.8 |
| 0017349 | 〃 | 4.6×25 | 18,000 | 0.7~1.8 |
| 0017350 | 〃 | 6.0×15 | 11,000 | 0.7~1.8 |

Table 8 Guaranteed Specifications

(continued)

| Part No. | Type | Column Size mm(I.D.)×cm(L) | Number of Theoretical Plates (TP/Column) | Asymmetry Factor |
|----------|-----------------------------|-------------------------------|--|---------------------|
| 0020017 | TSKgel Super-Phenyl | 2.0×5 | 3,000 | 0.8~1.8 |
| 0020018 | ∕ | 2.0×10 | 6,000 | 0.7~1.6 |
| 0018277 | ∕ | 4.6×5 | 8,000 | 0.8~1.6 |
| 0018278 | ∕ | 4.6×10 | 16,000 | 0.8~1.6 |
| 0007147 | TSKgel Silica-60 | 4.6×25 | 6,000 | — |
| 0006730 | ∕ | 21.5×30 | 6,000 | — |
| 0007146 | TSKgel Silica-150 | 4.6×25 | 6,000 | — |
| 0006723 | ∕ | 21.5×30 | 6,000 | — |
| 0007127 | TSKgel NH ₂ -60 | 4.6×25 | 6,000 | — |
| 0007128 | TSKgel OH-120 | 4.6×25 | 6,000 | — |
| 0007190 | TSKgel TMS-250 | 4.6×7.5 | 1,500 | — |
| 0013352 | TSKgel OligoDNA RP | 4.6×15 | 7,000 | — |
| 0013353 | ∕ | 7.8×15 | 7,000 | — |
| 0014005 | TSKgel Octadecyl-NPR* | 4.6×3.5 | 1,000 | 1.0~2.0 |
| 0018754 | TSKgel Octadecyl-2PW* | 2.0×15 | 5,000 | — |
| 0017500 | ∕ | 4.6×15 | 6,000 | — |
| 0017501 | ∕ | 6.0×15 | 6,000 | — |
| 0018755 | TSKgel Octadecyl-4PW* | 2.0×15 | 2,000 | 0.8~1.6 |
| 0013351 | ∕ | 4.6×15 | 2,000 | 0.8~1.6 |
| 0016257 | ∕ | 21.5×15 | 2,000 | 0.8~1.6 |
| 0017794 | TSKgel Enviropak G1* | 6.0×15 | 5,000 | 0.9~1.6 |
| 0020011 | TSKgel VMpak-25* | 2.0×5 | 1,000 | 0.7~1.6 |
| 0020012 | ∕ | 2.0×15 | 3,000 | 0.7~1.6 |
| 0018756 | TSKgel Phenyl-5PW RP* | 2.0×7.5 | 400 | 0.8~1.8 |
| 0008043 | ∕ | 4.6×7.5 | 500 | 0.8~1.6 |
| 0016260 | ∕ | 21.5×15 | 1,000 | 0.8~1.8 |
| 0014006 | TSKgel Phenyl-5PW RP Glass* | 5.0×5 | 400 | — |
| 0014007 | ∕ | 8.0×7.5 | 700 | — |

Note : The columns with the symbol "*" are of polymer-based gel and the others are of silica-based gel.

14. Column Cleaning Solutions

14-1 Silica based columns

- (1) TSKgel ODS-80T_M, TSKgel ODS-120A, TSKgel ODS-120T, TSKgel OligoDNA RP, TSKgel Super-ODS, TSKgel Super-Octyl, TSKgel Super-Phenyl, TSKgel ODS-100S, TSKgel ODS-140HTP 2.3μm, TSKgel Octyl-80T_S, TSKgel CN-80T_S, TSKgel TMS-250

- Sample property : Hydrophobic compounds

Water soluble organic such as 70%~100% acetonitrile and methanol in aqueous buffer

- Sample property : Basic compounds

Acidic solutions containing water soluble organic such as 70%~100% acetonitrile and methanol

- (2) TSKgel Silica-60, TSKgel Silica-150, TSKgel OH-120

Mixed solvents of organics such as ethanol and hexane (50/50 vol%), or

isopropyl alcohol and hexane (50/50 vol%)

(3) TSKgel NH₂-60

Water soluble organic such as 70%~100% acetonitrile and methanol in aqueous buffer

14-2 Polymer based columns

- 0.1 mol/L ~ 0.2 mol/L NaOH solution
- 20%~40% acetic acid solution
- Mixed solvent of 0.1 mol/L ~ 0.2 mol/L NaOH with organic such as acetonitrile (40/60 vol%); Inject several times using an injection valve with a 0.2 mL ~ 1.0 mL injection loop



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