Rev. BU030011

TOYOPEARL<sup>®</sup> lon Exchanger

650 Series TOYOPEARL GigaCap<sup>®</sup> S-650M TOYOPEARL GigaCap<sup>®</sup> Q-650M TOYOPEARL GigaCap<sup>®</sup> CM-650M

FOR IN VITRO USE ONLY / FOR PACKED-BED USE ONLY

# INSTRUCTION MANUAL



# **Safety Precautions**

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

# [Notation Conventions]

| Notation | Explanation  |  |
|----------|--|--|
|          | Indicates a potentially hazardous situation which could result in death or serious injury. |  |
|          | Indicates a potentially hazardous situation which could result in injury.                  |  |

#### 

#### Keep away from fire

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

#### 

| Use only in well-ventilated areas<br>In case of insufficient ventilation, flammable and toxic solvents can cause fire, explosion, or<br>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 in 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 as intended  |  |
| This product is for separation and purification. Do not use for any other purpose.   |  |
| Make sure compounds are safe   |  |
| Check that obtained compounds and solutions after separation and purification are safe.  |  |
| Proper disposal  |  |
| Dispose in accordance with local laws and regulations.   |  |
|  |  |
| NOTE   |  |
| NOTE   |  |

Keep this manual with the product for future reference.

# Precautions: Shipping Solvents

TOYOPEARL<sup>®</sup> Ion Exchangers are shipped in 20 % aqueous ethanol.

| Inhalation                                     | <ul> <li>Move the person to an area with fresh air.</li> <li>Immediately rinse the mouth with plenty of water.</li> <li>Call for medical attention immediately.</li> </ul>  |
|--|---|
| Skin exposure                                  | $\cdot$ Wash exposed area with plenty of soap and water.  |
| Eye exposure                                   | <ul> <li>Open eyes as wide as possible and rinse with clean water for at least 15 minutes.</li> <li>Call for medical attention immediately.</li> </ul>  |
| Ingestion                                      | Wash the mouth with plenty of water and immediately call for medical attention.   |
| Ventilation                                    | <ul> <li>Provide adequate air ventilation to keep organic<br/>vapor concentrations below approved level.</li> </ul>   |
| Container<br>handling                          | Container may break if not handled with care.   |
| Wear<br>appropriate<br>protective<br>equipment | <ul> <li>Use solvent-resistant gloves and protective eye gear<br/>when using this product. Use of gas mask, additional<br/>protective clothing or rubber boots could be appropriate<br/>when handling this product.</li> </ul>        |
| Hazardous<br>substance<br>storage              | <ul> <li>If any flammable solvents are used for shipping or<br/>storage, keep away from fire and open heat.</li> </ul>  |
| Storage<br>temperature                         | $\cdot$ Avoid storing this product at very low temperatures (<0 $^\circ\text{C}$ ) to prevent product from freezing.  |
| Disposal<br>methods                            | <ul> <li>Follow local guidelines for disposal. This product<br/>can be incinerated safely.</li> </ul>   |
| General considerations                         | <ul> <li>Please pay attention to all safety precautions with<br/>respect to the handling and storage of this product.</li> </ul>  |
|  | Skin exposure<br>Eye exposure<br>Ingestion<br>Ventilation<br>Container<br>handling<br>Wear<br>appropriate<br>protective<br>equipment<br>Hazardous<br>substance<br>storage<br>Storage<br>temperature<br>Disposal<br>methods<br>General |

# Precautions: TOYOPEARL<sup>®</sup> Brand Chromatographic Media

| -                 |  |  |
|-------------------|--|--|
| First Aid         | Inhalation                                     | <ul> <li>Move the person to an area with fresh air.</li> <li>Rinse the mouth with plenty of water immediately.</li> <li>Call for medical attention immediately.</li> </ul>   |
|                   | Skin exposure                                  | $\cdot$ Wash exposed area with plenty of soap and water.   |
|                   | Eye exposure                                   | <ul> <li>Open eyes as wide as possible and rinse with clean water<br/>for at least 15 minutes.</li> <li>Immediately call for medical attention.</li> </ul>   |
|                   | Ingestion                                      | Rinse the mouth with plenty of water and call for medical attention immediately.   |
| Handling<br>and   | Ventilation                                    | Provide adequate air ventilation to keep organic vapor concentrations below approved level.  |
| Storage           | Container<br>handling                          | Container may break if not handled with care.  |
|                   | Wear<br>appropriate<br>protective<br>equipment | <ul> <li>Use solvent-resistant gloves and protective eye gear when<br/>using this product. Use of gas mask, additional protective<br/>clothing or rubber boots could be appropriate when handling<br/>this product.</li> </ul>   |
|                   | Hazardous<br>substance<br>storage              | <ul> <li>If any flammable solvents are used for shipping or storage,<br/>keep away from fire and open heat.</li> </ul>   |
|                   | Fire<br>precautions                            | Do not expose the chromatographic resin to fire or open heat sources.  |
| Waste<br>Disposal | Disposal<br>methods                            | • This product can be incinerated or buried for easy disposal.<br>See below for additional precautions.  |
|                   | General considerations                         | Please pay attention to all safety precautions with respect to the handling and storage of this product.   |
|                   | Disposal<br>precaution                         | <ul> <li>Dispose in accordance with local laws and regulations.<br/>This product can be incinerated safely.</li> <li>Assure that appropriate countermeasures is taken when<br/>incinerating TOYOPEARL GigaCap<sup>®</sup> Q-650M.<br/>Exhausts may contain nitrogen oxides.</li> <li>Appropriate sulfur exhaust emission precaution should<br/>be taken for TOYOPEARL GigaCap<sup>®</sup> S-650M.</li> </ul> |

TOYOPEARL<sup>®</sup> products contain combustible packings based on a methacrylate polymer

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# 1. Introduction

TOYOPEARL GigaCap<sup>®</sup> series lon exchanger has been specifically designed for packed-column use, and is based on TOYOPEARL<sup>®</sup> HW-65 (650 Series, Protein Exclusion Limit 5 × 10<sup>e</sup>), which are the porous and spherical polymers.

They have the following features.

\* The change of gel volume is negligible in buffer with various pH or salt concentration.

\* Applicable to fast flow rate on column chromatography.

\* Resistant to microorganism

\* Applicable to HPLC system.

(Products Line-up)

| 650 Series | Strong Cation | TOYOPEARL GigaCap <sup>®</sup> S-650M  |
|------------|---------------|--|
|            | Strong Anion  | TOYOPEARL GigaCap <sup>®</sup> Q-650M  |
|            | Weak Cation   | TOYOPEARL GigaCap <sup>®</sup> CM-650M |

\* Note(Particle sizes) For 650 Series Medium 50-100  $\mu$  m

# 2. Procedure for Chromatography

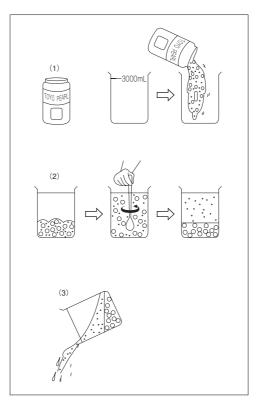
# 2-1 Removal of Fines

- (1) Transfer 500 mL of gel into a beaker of 3000 mL
- (2) Add distilled water up to 2000 mL in the beaker, stir and leave them until the gel precipitates.

Note: The necessary standing times of the gel with different particle sizes are as follows.

\* Medium Grade (GigaCap series) : 30-45 min.

- (3) Discard the supernatant (containing fines) by decantation.
- (4) Repeat the process (2) and (3) three or more time.



Removal of Fines

#### 2-2 Cleaning

TOYOPEARL Ion Exchangers are packed with 20 % aqueous ethanol.

The washing of the gel is necessary prior to use.

Pour the gel slurry on a glass filter and wash with distilled water of three times of the gel volume.

#### 2-3 Preparation of Gel Slurry and Packing

After removing fines from the gel by decantation, wash the gel with packing solvent should be used with the highest salt concentration in the used eluents, then transfer the gel into a beaker and add the packing solvent so as to make ca. 30-50 %(V/V) slurry.

The packing method under pressure (0.05-0.3 MPa) is desirable.

In this case a pump and reservoir are necessary for the packing.

Usually the flow rate of packing is two times faster than that of operation.

The gravitational packing method is often applied as conventional one.

In this case the pressure is desired to be as large as possible.

#### 2-4 Equilibration

After packing, the column should be equilibrated with 3 to 5 column volume of buffer.

#### 2-5 Elution

Adsorbed sample can be eluted by increasing of salt concentration up to 1 mol/L or change of pH in buffer.

#### 2-6 Regeneration

The gel can be regenerated by the following procedure.

#### 2-6-1 Batch method

Pour the gel into a beaker and the cleaning solvent in it, and stir and leave them until the gel precipitates, then discard the supernatant by decantation.

Repeat this process 2 or 3 times.

▲ Caution : The extremely severe cleaning method that was described below, will use HCl solution. Please note that some kind of protein will be aggregated in acidity condition.

TOYOPEARL GigaCap® S-650M, TOYOPEARL GigaCap® CM-650M

\* General cleaning

At first wash the gel with 0.5-1.0 mol/L NaCl solution by the procedure mentioned above, and then equilibrate the gel with buffer.

\* Severe cleaning

Wash the gel with 0.1-0.5 mol/L NaOH followed by washing 0.1-0.5 mol/L NaCl solution. Then equilibrate the gel with buffer.

\* Extremely severe Cleaning

Wash the gel with 0.1-0.5 mol/L NaOH, then water, then 0.1-0.5 mol/L HCl, then 0.1-0.5 mol/L NaCl, followed by washing with buffer.

#### TOYOPEARL GigaCap<sup>®</sup> Q-650M

\* General cleaning

At first wash the gel with 0.5-1.0 mol/L NaCl solution by the procedure mentioned above, and then equilibrate the gel with buffer.

\* Severe cleaning

Wash the gel with 0.1-0.5 mol/L NaOH followed by washing 0.1-0.5 mol/L NaCl solution. Then equilibrate the gel with buffer.

\* Extremely severe cleaning

Wash the gel with 0.1-0.5 mol/L HCL, then water, then 0.1-0.5 mol/L NaOH, then 0.1-0.5 mol/L NaCI, followed by washing with buffer.

#### 2-6-2 Column Method

The gel in a column can be regenerated easily by flowing the cleaning solvents on the column.

The procedure and the solvents for the cleaning are just same as that of Batch Method.

#### [Advantages of Column Method]

| * Simple Handling      | Taking out of the gel from the column and repacking of |
|------------------------|--|
|                        | the gel into the column are not necessary.             |
| * Good Reproducibility |  |
| * Quick Cleaning       | By applying a pump the cleaning time becomes shorter   |
|                        | than that by Batch Method.                             |
| * Effective Cleaning   | The gel can be regenerated well with a small amount of |
|                        | solvents compared with Batch Method.                   |

# 3. Storage

The gel should be stored with 20 % aqueous ethanol at ambient (4–35  $^\circ C$  )

# 4. Remarks

#### 4-1 Removal of Fines

As described in Section 2, remove fines before use.

When the removal of fines is not performed completely, micro-particles leak from column during chromatography.

Leak of micro-particles, however, would be stopped in short time.

#### 4-2 Clogging of Filter

Increasing of pressure-drop or decreasing of flow-rate is caused by clogging of filter.

In this case, take out the gel from the column, clean the fitting and repack the gel into the column.

#### 4-3 Adsorption of Protein

When protein is not adsorbed well to column with the initial buffer, sample should be dialyzed or desalted.

#### 4-4 Packing Method

Pack the gel to column by pressure-packing method.

Packing of column by suction method can not be recommended for the column more than 10 cm long.



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