Proteonavi

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Features

·Excellent acidic durability

·Minimal protein adsorption and minimal sample loss

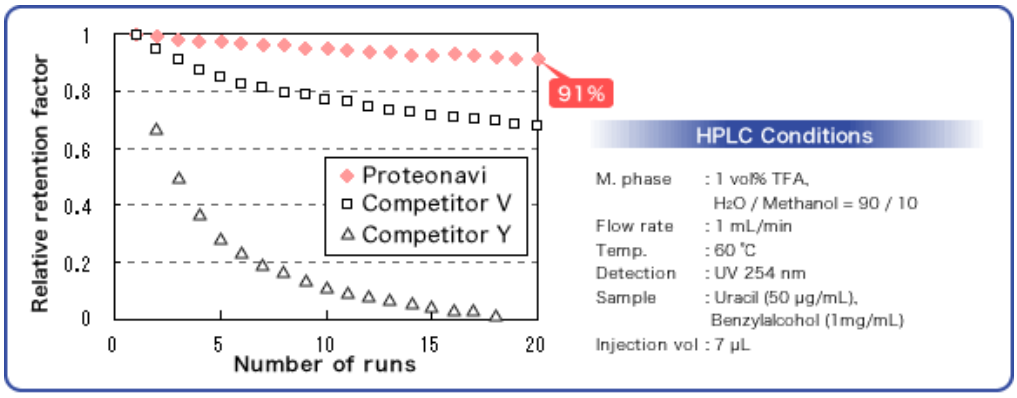
·Easy to shift from analytical to preparative size

·Also available in worldwide

Adsorption to a stationary phase is one of the most common limiting factors in protein separation in reversed-phase mode. It is generally understood that the irreversible adsorption is caused by denaturing of protein in the hydrophobic phase or a coulombic interaction with silica, a chromatographic support. Proteonavi has overcome the problem by introducing the short four- carbon structure on the silica surface with a unique chemistry. Its synthetic process has already been established for even a large industrial-scale purification.

Outstanding acidic durability

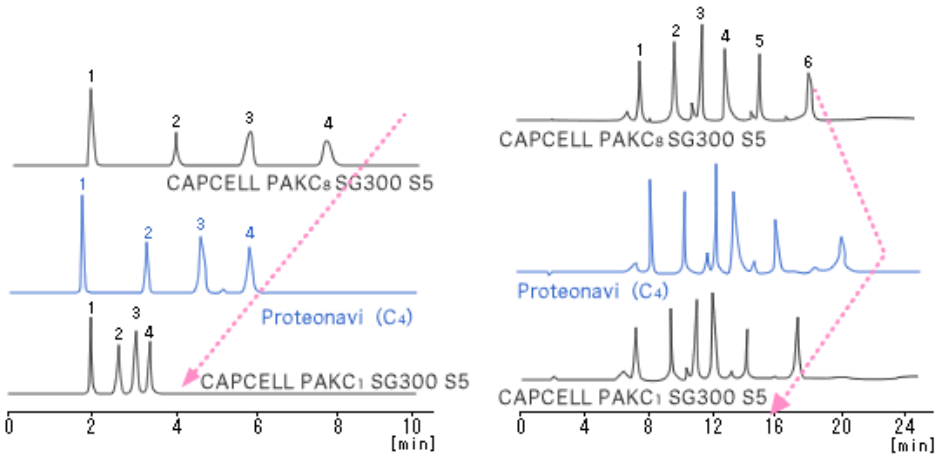
Acidic hydrolysis is the major cause of loss in performance in reversed phase. Proteonavi's durability under acidic conditions was proven by the accelerated test using 1vol% of trifluoroacetic acid (TFA), a concentration one order of magnitude higher than those used for mobile phases for common protein separations.
Sequence of process: After thermal equilibration of column, start the pump. Sixty minutes later, run the sample and record its retention time. Repeat the sequence in every 60 minutes and observe the loss of retention. (For HPLC condition, see below.)



Acid durability test

Specific Retention for Proteins

While their retention times of small neutral compounds are supposed to be correlated to amount or length of alykyl chains of stationary phase, that of protein is, in general, governed not only by hydrophobic interaction, but by hydrophilic or ionic interactions. Proteonavi is designed to show large retention specifically for proteins, by precisely controlling its synthetic process.



HPLC Conditions

Column size : 2.0 mm i.d. x 150 mm
Mobile phase : CH_3CN / H_2O = 40 / 60
Flow rate : 200 μL/min
Temp. : 40 °C
Detection : UV 254 nm
Sample : 1. Uracil, 2. Methyl benzoate, 3. Toluene, 4. Naphthalene

HPLC Conditions

Column size : 2.0 mm i.d. x 150 mm
Mobile phase : A) 0.1vol% TFA / H_2O
Flow rate B) 0.1vol% TFA / CH_3CN
Temp. B 20%(0min) → 70%(25min) Gradient
Detection : 200 μL/min
Sample : 40 °C
: UV 220 nm
: 1. Ribonuclease A, 2. Cytochrome C, 3. Lyzozyme, 4. B.S. albumin, 5. Myoglobin, 6. Ovalbumin

Specific Retention for Proteins

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Proteonavi is provided with packing material made of totally porous spherical silica bonded with C4. The epoch-making packing material integrates the high separation performance of proteins and peptides, pressure resistance of silica packing material, and minimal protein adsorption.

1. Handling the Column

1. Handle the column with great care. A strong shock may cause damage.
2. Attach or detach the column when the pressure gage indicates zero.
3. The maximum column operating pressure is 20 MPa.

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2. Attaching the Column

1. The column joint is of the male nut type for tubing of 1/16 inch OD. Check that the tubing joints of the system fit correctly and that the ferrule tips are deeply inserted into the joints. (See Fig. 1.)
2. Before attaching the column, replace the liquid in the system with the mobile phase to be used. Note the replacement procedure to avoid salting out. The shipment solvent is described in the column report enclosed with the column.
3. Attach the column according to the direction of the arrow.

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3. Analysis

3-1. Mobile Phase

1. All solvents acceptable for the conventional chemically bonded silica columns can be used.
2. The acceptable pH range for Proteonavi is 2 to 10.
3. After full degassing, filtrate the mobile phase using a membrane filter 0.45 µm or smaller to remove dust. A 2-µm filter is used at the column inlet. To prevent foreign matter from clogging the column inlet filter, we recommend using a line filter.
4. The mobile phase stated in the report is sealed in a new column. To change to a mobile phase containing inorganic salt, note the replacement procedure.
5. To prevent column deterioration, avoid the following:
   •Frequent change of the mobile phase composition or direct change to a mobile phase of low compatibility
   •Rapid change in column inlet pressure
   •High column pressure due to the use of a high-viscosity mobile phase
   •Prolonged water flow

3-2. Preparing a Sample Solution

1. Dissolve the sample in a solvent of the same composition as the eluate wherever possible.
2. Using a solvent with strong dissolving power may lower the separation efficiency or cause the sample to precipitate at the column head.
3. If there is insoluble matter remaining in the sample solution, filtrate the solution using a filter 0.45 µm or smaller.
4. The pH of the sample solution should be set in acceptable pH range for packing material.

3-2. Notes on Analysis

1. Regarding the guard column: Use a column of the same packing material as the main column. If a guard column of different packing material or the chemically bonded silica column of a different manufacturer is used, a separation profile may not be as expected.

2. After gradient analysis is performed, replace the column with eluent containing high content of organic solvent. See 4. for storing.

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4. Storing the Column

1. Seal the column with the accessory plug and store it in a cold place where there is little temperature fluctuation.
2. For storage, replace the mobile phase with a solution of acetonitrile and water having the same composition as the mobile phase and then fill it with acetonitrile. Note the replacement procedure to avoid salting out.
3. Avoid using 100% water to rinse a column.

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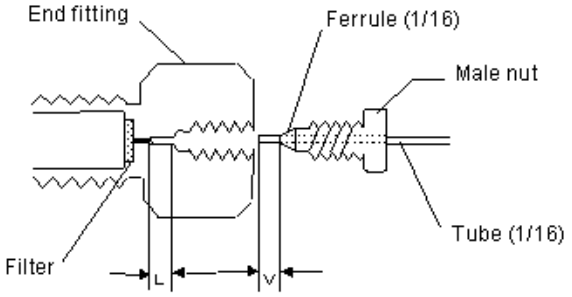
5. End Fittings

1. An analytical column of up to 6-mm ID uses a filter-embedded end fitting as shown in Fig. 1. The filter cannot be changed alone. If the filter is clogged or the column pressure is high, replace the end fitting. See Table 3 for the replacement parts and repair items.
2. See Fig. 1 for the column connection. If the tubing is inappropriate, especially if a tube for a different type of column is used, the length after the ferrule tip (V in Fig. 1) is often different from the end fitting length L, and a problem may occur.
   If L is greater than V, dead volume may be generated and cause peak broadening or tailing or deterioration of separation performance.
   If L is smaller than V, liquid may leak because of inadequate ferrule adhesion.
   Therefore, we recommend replacing the ferrule together with the column.
   *If the column is replaced frequently, the male nut may crush the ferrule and liquid may leak. Since tightening the nut too much may cause its head to come off, replace the ferrule at an early stage.



【Fig. 1】 Column connection

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Proteonavi is shipped after a strict performance check. However, if you should find any defect, please contact your dealer or Shiseido for replacement. Note that Shiseido does not warrant the product against column life or deterioration caused by the failure to follow the above handling instructions. Ten or more days after reception by the customer, Shiseido will assume that the product was delivered in good condition, and will not accept a later replacement request.

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