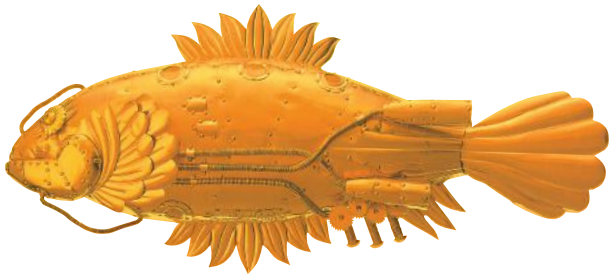
Shodex™



HPLC Columns

MANUAL

Protein LW-803

SHOWA DENKO EUROPE

Shodex HPLC Columns
Europe, Middle East, Africa, Russia

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Operation Manual

Shodex™ PROTEIN LW-803

(Please read this manual carefully before using the column to ensure performance and life.)

1. Introduction

The packed column of Shodex PROTEIN LW-803 is designed to be used in high performance gel filtration chromatography for precision separation of proteins and/or enzymes. This product is proper to analyze protein of several hundred thousand molecular weight by regulating pore size.

2. Handling Instructions <Important>

Caution! Please consult the SDS for the reagents and solvents used with the columns for health concerns caused by acute exposure due to leakage from the column or adjoining tubing.

Attention! Use the column within the regular range of flow rate, pressure and temperature. The column performance may deteriorate when it is handled beyond the permissible range even for a short time. See the Section 4. Usable Conditions for the permissible ranges.

3. Specifications

Product code	Product name	Size (mm)		Particle size (µm)	Exclusion limit (Protein)	Molecular range	Plate Number (TP/column)
		I.D.	Length				
F6989303	PROTEIN LW-803	8.0	300	3	*(1,000,000)	10,000 - 700,000	≥ 12,000
F6700133	PROTEIN LW-G 6B	6.0	50	3	-	-	Guard column

* estimated value

Packing material : Porous silica particle bound with hydrophilic group polymer

Column material : SUS316

Screw type: Unified Thread Standard No. 10-32 UNF

Shipping solvent : H_2O

4. Usable Conditions

Product name	Flow rate (mL/min)		Max pressure (MPa)	Temperature range(ºC)	pH range
	Standard	Max			
PROTEIN LW-803	1.0	1.5	15.0	4 - 45	3.0 - 7.5
PROTEIN LW-G 6B			-	4 - 45	

5. System Clean up

Clean up the LC system including the injector and the sample loop by switching the valve, and flow the eluent before column installation.

Attention! The previous eluent used for analyses in the system may damage the column, if it is not compatible with the column.

Attention! When replacing nonpolar solvent to water, replace first with methanol and then replace with water. When replacing buffer solution to acetonitrile/water, replace first with 100% water and then repalce with eluent. Substances adsorbed in the pump and tubing may not be compatible with the column.

6. Column Installation

Install and use the column with the flow through the column matching the flow direction arrow on the column tag. Set the flow rate at 0.5 mL/min, and connect the column. Flow at the low rate until the column temperature increases to the setting temperature, and then increase the flow rate to the analytical condition.

Attention! The column should always be installed in the manner above, for safe and effective operation.

7. Eluents

1) Thoroughly degas the solvent that is to be used as the eluent, and pass through a 0.45µm membrane filter.

Attention! Filter the eluent with a 0.45µm membrane filter to prevent chromatogram noise and column performance deterioration by small particles or undissolved materials.

2) Phosphate buffer solution, tris hydrochloric solution, acetate buffer solution are generally used as the eluent.Salts such as Na_2SO4 K_2SO4, (NH4)2SO4 are usually added to the above mentioned buffer solutions.The suitable salt concentration is 0.1 to, 0.3M.

3) Use the column within the eluent pH range.

Attention! The pH range of the eluent should be 3.0 to 7.5 range.

4) Acetonitrile and methanol can also be used as the eluent.The concentration range should be between 0 and 100%.

Attention! Confirm the salt precipitation when polar organic solvents are mixed with buffer solution.

5) Protein denaturation reagents (ex. urea or guanidine hydrochloride) and surfactants (ex. SDS or Brij) can be used.

Attention! Since the eluent containing urea or guanidine has high viscosity, keep the flow rate at 0.5mL/min or less.

Attention! Since protein denaturation reagents or surfactants may remain in the column, use the same eluent condition.

8. Sample Preparation

1) Remove extraneous matter or gels from the dissolved sample by passing it through a 0.45µm filter.

2) Dissolve the sample in the same solvent that is to be used as the eluent.

9. Column Cleaning

Elution characteristics of a column may change considerably after long, repeated usage, due to the accumulation of pollution components on the packing material from the LC system or the sample. The cleaning procedures outlined below may be used. Clean the guard column and analytical column separately by flowing the cleaning solution in the opposite direction of the arrow on the column tag. The applied flow rate should be lower than half of standard flow rate. The volume of the cleaning solvent required is 5 to 10 times the column volume.

1) In case where protein has been adsorbed :

Use a solvent containing 0.5M salt or phosphate buffer (about pH3.0).

2) In case where a hydrophobic substance has been adsorbed :

Use a solvent containing 10 to 20% acetonitrile or methanol.

Attention! Confirm the salt precipitation when polar organic solvents are added to the eluent.

Attention! In case where protein has been adsorbed in column, protein may precipitate by flowing a high concentration polar organic solvent.

10. Column Inspection

Column inspection method is described in Certificate of Analysis (CoA).

Attention! Analyze by inspection condition on the Certificate of Analysis and Confirm column performance after purchasing and in each used.

11. Attention

1) Do not remove the end fittings of the column to prevent performance deterioration and for safety reasons.

2) Do not make a strong impact on the column: such as hitting or dropping on the floor.

3) Replace the solvent in the LC system with the eluent to be used before connecting the column.

4) Connect the column so that the flow direction corresponds to the arrow mark on the tag.

5) When the column is not used for two weeks or more, replace the in-column solvent with the shipping solvent, remove it from the LC system, close each end with a stopper, and store it at controlled room temperature.

6) Contact Shodex website (http://www.shodex.com/) or Shodex partners regarding produce

and analysis applications.

12. Warranty (Ver. 3)

1) Showa Denko K. K. warrants that the ShodexTM Column, at the time of delivery to the user, will conform to the specification of the attached Certificate of Analysis, if the Shodex Column is used in accordance with the attached operating manual. The foregoing warranty is exclusive and is in lieu of all other warranties with respect to the Shodex Column, whether written, oral, implied, statutory or otherwise. No warranties by Showa Denko K. K. are implied or otherwise created, including, but not limited to, the warranty of merchantability and fitness for particular purposes.

2) Any claim of inconformity to the specification must be notified to Showa Denko K.K. within ten (10) days after delivery to the user. User's exclusive remedy and Showa Denko K.K.'s exclusive liability for such claim are limited to the replacement of the Shodex Column in question. In no event is Showa Denko K.K. liable for any indirect, incidental or consequential damage arising out of in connection with the Shodex Column, whether or not such damage is allegedly based on breach of warranty, negligence or otherwise.

3) No warranty is made in any of the following cases:

(1) If the Shodex Column is not used in accordance with the operating manual

(2) If the Shodex Column is remodeled by anyone other than person or firm designated by Showa Denko K.K.

(3) If the Shodex Column is disposed of

(4) If the Shodex Column is resold by the user without giving prior written notice to Showa Denko K.K.

(5) If the performance of the Shodex Column is not conform to the specification of the attached Certificate of Analysis due to any of the reasons below:

(a) Computer virus

(b) Impurities contained in the sample, reagent, gas air or cooling water provided by the user

(c) Breakdown or malfunction of equipment, apparatus or component used in combination with the Shodex Column

(d) Force majeure such as fire, earthquake, flood, other natural disaster, crime, riot, act of terrorism, war or radioactive contamination

4) In no event is Showa Denko K.K. liable for

(1) the results of analyses or preparations using the Shodex Column or any portion of the same, including, but not limited to, the reliability, accuracy, efficacy and safety of said results, and

(2) the occupational hazard in the use of the Shodex Column, whether or not such use is made in accordance with the attached Conditions for use.

5) The Shodex Column is for laboratory use only. It must not be used for clinical diagnosis. Showa Denko K.K. is not liable for any use of the Shodex Column except laboratory use.