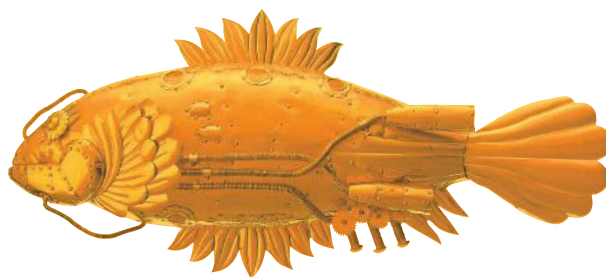


Shodex™



HPLC Columns

MANUAL

RSpak DE-413

SHOWA
DENKO
EUROPE

Columns manufactured by Showa Denko K.K Japan
Made in Japan

Shodex HPLC Columns
Europe, Middle East, Africa, Russia

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Operation Manual

Shodex™ RSpak™ DE-413

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

1. Introduction

DE-213, DE-413 and DE-413L of the Shodex RSpak DE-413 series are high-performance HPLC columns designed for absorption/partition chromatography. These columns are highly effective in analyzing samples, such as foods, drugs and biological substances, which have a wide range of polarity. The columns show excellent performance.

2. Specifications

Packing material: Polymethacrylate

	DE-213	DE-413	DE-413L
Column size	2.0 mm ID x 150 mm length	4.6 mm ID x 150 mm length	4.6 mm ID x 250 mm length
Theoretical plate number	>8,000	>11,000	>17,000
Maximum pressure	12.0 MPa	15.0 MPa	18.0 MPa
Maximum flow rate	0.4 mL/min.	1.2 mL/min.	
Usable temperature	Room temperature ~ 60°C		
Usable eluent	Mixture of water or buffer and water soluble organic solvent such as methanol, ethanol, 2-propanol, acetonitrile and THF		
Usable buffer	Buffer of pH 2 ~ 12 such as phosphate buffer and acetic buffer		
Usable salt concentration	<0.5 %		
In-column solvent	Water/Acetonitrile = 50/50		
Endfitting	Internally-threaded type, No. 10-32 UNF		
Column material	SUS 316		

3. Analytical conditions for test chromatogram

Sample: Di-n-butyl ketone 0.6 v/v% in 50% Acetonitrile.
Injection volume: 5 μ L (DE-213), 10 μ L (DE-413), 20 μ L (DE-413L).
Eluent: Water/Acetonitrile = 50/50.
Flow rate: 0.2 mL/min. (DE-213), 1.0 mL/min. (DE-413, DE-413L).
Detector: UV (262nm).
Column temperature: 40°C.

4. Recommended analytical conditions

Eluent: Mixture of water, phosphate buffer or acetic buffer and acetonitrile or methanol.
Flow rate: 0.05-0.3 mL/min.(DE-213), 0.6-1.0 mL/min. (DE-413,DE-413L).
Column temperature: 40 °C.

5. Change of eluent

If eluent need to be changed, first pump new eluent into the column at a flow rate of 0.05 mL/min. for about 40 minutes for DE-213, 0.2mL/min. for about 40 minutes for DE-413 column and 60 minutes for DE-413L. If the concentration of the organic solvent in the new eluent is equal to that the eluent to be changed, pass the new eluent through the column at a flow rate of 0.1mL/min. for 20 minutes about DE-213 and 0.5mL/min. for 20 minutes for DE-413 and DE-413L. If not, send a salt-free solution through before change of the new eluent.

For example, in case of DE-413 to change a mixture of 50 mM phosphate buffer (pH3.1) and acetonitrile (at a ratio of 48/52) with 50 mM phosphate buffer (pH3.1), the procedures are below. (It is a similar procedure of IIE-213, DE-413 and DE-413L)

- 1) Pump a mixture of water and acetonitrile (at a ratio of 48/52) into the column at a flow rate of 0.5mL/min. for 20 minutes.
- 2) Then pass water through the column at a flow rate of 0.2mL/min. for 40 minutes.
- 3) Finally, allow 50mM phosphate buffer (pH3.1) to flow through the column at a flow rate of 0.5mL/min. for 20 minutes.

For DE-213 and DE-413L, the difference in the concentration of organic solvent in the eluent to be changed and that in the new eluent should not be more than 50%. If the difference is more than 50% and the eluent needs to be changed, it should be changed in steps so that the difference in the concentration at each change is less than 30%. We recommend that they are used without changing the eluent.

6. Regeneration

If impurities adsorbed on the packing material have to be removed, pump 50mL of acetonitrile or methanol into the column at a flow rate of 0.1mL/min. (DE-213), 0.5mL/min. (DE-413,DE-413L).

7. Storage

- 1) After completing analysis, keep pumping eluent into the column at a flow rate of 0.05mL/min. (DE-213), 0.2mL/min. (DE-413,DE-413L) until the column is cooled down to room temperature.
- 2) Disconnect the column from the chromatograph and cap both ends of the column to prevent the eluent from leaking out. Then, pack the column in the same way as when it was delivered from the manufacturer and store it in a room where the temperature fluctuates little.
- 3) When the column is not to be used for a long time, fill it with completely degassed 50% acetonitrile before storing it. For replacement of in-column solvent, please refer to the "5. Change of eluent".

8. Caution

1. Do not store the column under strong acidic or strong alkaline conditions. If the eluent contains chlorine ions, the pH value must be over 4.0.
2. Do not bend the column or cause impact on it.
3. Do not change the column pressure or the flow rate abruptly when a liquid chromatograph is in use.
4. Under no circumstances should the end-fittings of the column be removed as doing so will cause deterioration of the column performance.

5. Install guard column immediately upstream of the main column to protect it from contamination by the sample. The guard column is installed to maintain the performance of the main column as designed for a long period of time and not to improve its resolving power.

9. Warranty

1) Showa Denko K. K. warrants that the Shodex 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 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 Instrument, 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 resold by the user without giving prior written notice to Showa Denko K.K.

(4) 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, rime, riot, act of terrorism, war or radioactive contamination

4) In no event is Showa Denko K.K. liable for (i) 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 (ii) 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 instrument 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 Instrument except laboratory use.