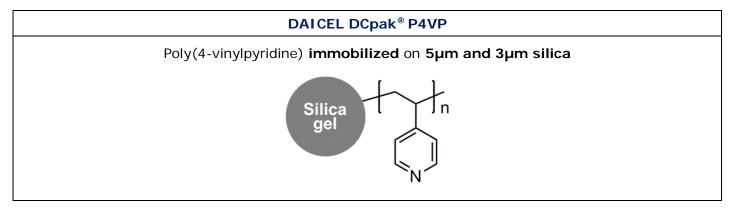




INSTRUCTION MANUAL FOR DAICEL DCpak® P4VP

Please read these instructions completely before using this column

Column Description



Shipping solvent: 100% Ethanol

Every column has been examined and quality control tested before shipping. Please refer to the Column Performance Report and test parameters for results.

CAUTION

The column is designed for 34.35MPa maximum pressure and for 30MPa daily pressure. Please use the column **neither** at a pressure over 30MPa **nor** at a temperature over 40 °C.

Please flush the residual solvent in the SFC instrument with a recommended mobile phase (see page 2) before connecting the column to the instrument. Please be sure to flush the auto-sampler, syringe, needle, and injection loop as well.

Operating Conditions

	50 x 2.1 mm i.d. 100 x 2.1 mm i.d. 150 x 2.1 mm i.d. 250 x 2.1 mm i.d. Analytical columns	50 x 3.0 mm i.d. 100 x 3.0 mm i.d. 150 x 3.0 mm i.d. Analytical columns	50 x 4.6 mm i.d. 100 x 4.6 mm i.d. 150 x 4.6 mm i.d. 250 x 4.6 mm i.d. Analytical columns	250 x 10 mm i.d. 250 x 20 mm i.d. 250 x 30 mm i.d. 250 x 50 mm i.d. Semi-prep/prep columns
Column Fittings	Waters			
Flow Rate Direction	As indicated on the column label			
Pressure Limitations*	30MPa (~ 305 kgf/cm ² or ~ 4350 psi)			
Temperature	0 to 40 °C			

*Pressure means the pressure at the column head, which is nearly equal to the pump pressure. The recommended back pressure regulator (BPR) setting is 8 – 20MPa. If the BPR setting is too low, an unstable chromatogram may result.

- > This column is not for chiral separations.
- > Do not attempt to disassemble the column.
- > This instruction sheet for DCpak® P4VP is not applicable to any other Daicel column.

Please contact your local Chiral Technologies office for further assistance before trying any solvents not mentioned below.

A – SFC Mobile Phases

	CO ₂ /co-solvent	
Composition	100/0 to 40/60	

- Methanol is typically used as a co-solvent. Ethanol, 2-propanol, ethyl acetate, THF, and dichloromethane can also be used.
- □ The eluotropic strength of the alcoholic co-solvents are methanol>ethanol>2-propanol if the same volume percentage is applied. This tendency becomes remarkable for a polar analyte.
- □ A higher co-solvent content results in a shorter retention time.
- □ A mixed co-solvent of the above organic solvents can also be applied. When an **aprotic** co-solvent is employed, the addition of alcohol in a small amount may improve peak shape.
- An increase of the co-solvent content increases the column head pressure. Pressure should not exceed 30 MPa.

B – Additives

- Initial method development can be performed without the addition of any additives. If needed, the recommended additives as illustrated in the table may help sharpen the peak shape.
- □ Typical concentration is 0.1 vol% of the total mobile phase. (e.g. use co-solvent containing 0.5% of additive if CO₂/ co-solvent ratio is 80/20 v/v).

Additive for basic	Additive for acidic
analyte	analyte
Diethylamine Ammonium formate Ammonium acetate	Trifluoroacetic acid
	~0.1 vol% of total
~0.1 vol% of total mobile phase	mobile phase

□ After a basic or acidic additive has been used, wash the column with more than 10 column volumes of mobile phase without additive, and then flush the column with ethanol.

Sample Preparation

□ The sample should be dissolved in the mobile phase co-solvent, i.e. methanol, ethanol, etc., and should be filtered through a membrane filter of approximately 0.5µm porosity.

Gradient and LC

- Analysis can be performed by gradient elution, however baseline instability may occasionally occur, in particular after using basic or acidic additives. Before performing a gradient analysis, complete a "test run" to verify baseline stability.
- While these columns were initially designed for use under SFC separation conditions, they can also be used under LC conditions in both Normal Phase (hexane/alcohol) and Reversed Phase modes. Please contact our technical assistance for suggested starting conditions.

Column Care / Maintenance

- □ After performing analyses which contain additives, it is good practice to flush the column with mobile phase which does not contain any additives. If removing the column from the system, flush with 100% ethanol or isopropanol first, and then remove the column following the notes below.
- Remove the column from the instrument ONLY after the inner pressure is completely released. Removing the column under a high inner pressure may cause hazards by rapid releasing CO₂, and can cause a deterioration of the column seal. Be sure to slowly loosen the connection to avoid possible release of CO₂.
- □ The column can be stored long term at ambient temperature.
- □ If reproducibility has been compromised, clean the column with more than 10 column volumes of ethanol at 1.0 mL/min. In addition, if the reproducibility has been compromised **AND** trifluoroacetic acid has been used repeatedly as an additive, it might be necessary to perform the column cleaning with 0.1vol% diethylamine in the mobile phase.

Operating this column in accordance with the guidelines outlined here will result in a long column life.

⇒ If you have any questions about the use of this column, or encounter a problem, contact:

- In the USA: <u>questions@cti.daicel.com</u> or call 800-6-CHIRAL
- In the EU: <u>cte@cte.daicel.com</u> or call +33 (0) 3 88 79 52 00
- In India: <u>chiral@chiral.daicel.com</u> or call +91 84 1866 0700 & 703

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