The Nucleonavi is specialty column designed exclusively for the analysis of nucleic acids and nucleic acid drugs, comprising monolithic silica, a single-piece full porous silica, as its base material. In addition to being metal free, Nucleonavi characteristics include high resolution that enables the discrimination of differences in base-chain lengths and constituent bases, separation at low pressure, and suppression to the absorption of nucleic acid sample.

To ensure stability and long product life, please read this manual carefully for correct use.

1. Column Handling

Except for the connector section, the columns are made of glass. Please take note of the following points.

1. Dropping or strong shock may cause damage. It is very dangerous. Handle with great care.
2. Attach or detach the column when the pressure gage indicates zero.
3. While the maximum acceptable pressure of the column is 20 MPa, use it at a normal operation pressure of below 15 MPa.
4. If any liquid mass forms inside the cover-glass (see Fig. 1), stop operation immediately. There is a possiblity that the glass rod may have ruptured. If the operation is continued, the glass cover might rupture and cause great danger.

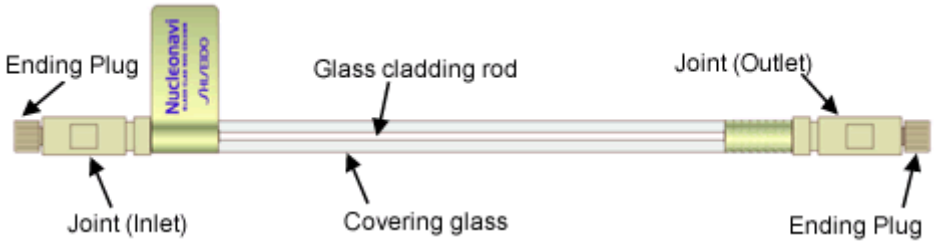

Fig. 1

2. Column Installation

1. The column joint is of a male-nut type for 1/16 inch OD tubing. Use tubing made of PEEK. Ensure that the tubing joints of the system fit correctly. Pay particular attention to ensuring that the ferrule tip is deeply inserted into the joint (See Fig. 2) .
2. Before attaching the column, displace the liquid in the system with the mobile phase to be used.
   * The shipment solvent is acetonitrile. When displacing it, beware of salting out and other reactions.
3. Install the column in the direction of the arrow on the column label.
4. When installing the column, remove the plugs on both ends, hold the joint (made of PEEK) securely on the inlet side of column and insert the PEEK tubing all the way to the bottom. Holding the cover-glass part or the joint (made of PEEK) on the outlet side when screwing in the tubing risks damaging the column.
5. To install the outlet side of column, run the liquid with the outlet side kept open until the mobile phase starts flowing out and the pressure stabilizes, and then stop the pump to install the part. When installing, as with the inlet side, hold the joint (made of PEEK) securely on the outlet side, insert the PEEK tubing to the bottom.

3. Analysis

3-1. Mobile phase

1. Applicable types of solvents are those of methanol/water and acetonitrile/water. For the measurement of nucleic acids or nucleic acid drugs, use a buffer solution containing an ion-pair reagent as the mobile phase. Incidentally, since the use of a different ion-pair reagent diminishes the reproducibility of data, please prepare a different column for evaluation by altering the ion-pair reagent.
2. Mobile phase should be used at a pH value in the vicinity of neutrality. Moreover, the alteration of pH will lead to diminished reproducibility or premature deterioration of the column.
3. Use mobile phase after filtering it (through a filter of 0.45 μm or less) to eliminate insoluble matter and dirt, and after degassing it.
4. With a view to preventing the clogging of column due to contamination with foreign matter, use of a liquid line filter is recommended.
5. Avoid the use of a 100% aqueous mobile phase. Otherwise, service life may be significantly shortened.

3-2. Sample preparation

1. For analysis, dissolve nucleic acids mobile phase or water.
2. Select a sample solution pH value in the vicinity of neutrality, as with the mobile phase.

3-3. Points to note in analysis operation

1. Ensure that the HPLC system to be used enables the use of 1 mm I.D. columns and has the capacity to run solution at a stable flow-rate of 50μL/min.
2. To minimize system dead volume using our NANOSPACE system, replace the tubing from injector to column and from column to detector with 0.13mm I.D. (red) PEEK tubing. (Product name: PEEK tubing 1/16 in. x 0.13mmi.d. x 3m Red, Product number: 2331)
3. When using our detector UV-VIS for detection, select our Semi-micro Cell (3μL) and adjust the time constant to RAPID.
4. For a mixer, use our low volume mixing column (50μL). (Product name: Low-volume mixing column, product number: 3451)
5. Terminate gradient analysis after displacing the mobile phase with one containing a larger fraction of organic solvents. For storage, consult 4. Storing the column

4. Storing the column

1. If the column is not to be used, displace the mobile phase with acetonitrile. In the displacement procedure, beware of salting out and the resultant rise of column pressure.
2. Seal the column with the accessory plug and store it in a cold place with little temperature fluctuation.
3. Avoid using 100% water to rinse the column.

5. About the column connection

For tubing, follow the instructions in Fig. 2. Inappropriate tubing, particularly when a tube used for a different type of column is used as is, the tip length beyond the ferrule (V in Fig. 2) may not match the length of the end fitting bore (L in Fig. 2), which leads to troubles.

Where L>V, dead volume occurs and may cause the broadening or tailing of peak, or limit separation performance.
Where L<V, inadequate sealing of the ferrule causes liquid leakage. Therefore, it is recommended that the ferrule be changed concurrently with the column change.

* After frequent column replacements the male-nut may present a crushed ferrule and cause liquid leakage. In such a case, tightening too hard may cause the head portion of the nut to break off. Therefore, replace the ferrule in good time. Moreover, for plumbing and ferrule, please use products made of PEEK. In particular, we recommend our push-screw integrating a male nut and a ferrule (rounded PEEK type, product number: 2223). Use of metallic tubing or ferrule may cause a leak.

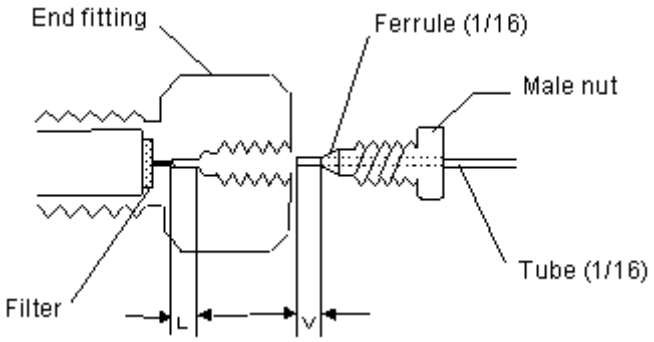

Fig. 2

While Nucleonavi columns undergo strict verification of performance before shipping, if any defect is identified, please inform us or our distributor of it.
However, please take note that we are not responsible for matters related to column service life or any problem caused by the failure to follow the above mentioned instructions for care and use.
Please note that we assume the product was delivered in good condition if no claim is made within ten days after receipt by the customer, and requsts for replacement submitted thereafter will not be accepted.