Packed Column for High-Performance Liquid Chromatography

# ProteoSil Instruction Manual

### 1 Introduction

Thank you for purchasing this product.

Please read this instruction manual carefully and use the product properly to ensure the best performance.

### 2 Handling Precautions

### 2-1 Precautions

- Do not drop or hit the column. Strong impacts may cause column degradation.
- The column provides high pressure resistance because it is packed using a high-pressure slurry method. However, in order to ensure long-term stable use, we recommend using the column at pressures not exceeding than those shown in the table below.

Particle diameter	Column size	Recommended pressure
1.9 µm	All sizes	80 MPa or less
3 µm HP	All sizes	50 MPa or less
3 - 10 μm	I.D. 0.3 to 50 mm	20 MPa or less

- · Be careful of sudden pressure fluctuations.
- · When removing the column, wait until the pressure gauge indicates 0.
- Please note that slow operation of the sample injection valve will cause sudden pressure changes at the column inlet.
- To the extent possible, dissolve the sample in a solvent of the same composition as the eluent (initial solvent for gradients).
- Injecting a large amount of sample dissolved in a solvent with a higher solubility than the eluent can result in reduced separating capacity and precipitation of the sample at the inlet of the column.
- To replace eluents that do not mix with each other (e.g., changing from a normal-phase type to reversed-phase type), an intermediate-polar solvent such as 2-propanol should be poured in between.
- At such a time, since alcohol-based solvents are subject to high pressures during feeding, adjust the flow rate while paying attention to the upper pressure limit of the column, and feed the solvent at a rate of at least 10 times the column capacity.
- If a peak that elutes quickly tails off, the cause may be due to dead volume. Check if the connection
  piping at the column joint is inserted all the way to the end.
- For piping to the injector and detector, select piping with an inner diameter and length suitable for the applicable column inner diameter and corresponding analytical system. The effect of piping is particularly significant when analyzing at low flow rates using semi-micro columns, etc.
- Pressure rise and peak cracking may be caused by clogging or contamination of the filter at the column inlet.
- Filter the eluent through a membrane filter of 0.45 µm or less before use.
- Filter the sample solution with a syringe filter of 0.45 µm or less before injecting.
- Column clogging can be prevented by using Cartridge E or Cartridge E 2.1x10 mm UHPLC.
- Ensure that the column is well balanced with the eluent before use.
   (Reversed-phase mode using HILIC or ion-pair reagents may take longer to stabilize.)
- Depending on LC/MS analysis conditions and the type of equipment, precipitates may be seen at the column outlet. Check the effect of precipitation before use.

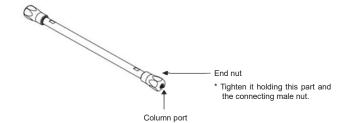
- efer to the table below for the applicable pH range and upper temperature limit of the column. (Refer to the GL Sciences website for the applicable pH range and upper temperature limit for columns not listed in the table.)
- Please note that exceeding the upper temperature limit can lead to premature deterioration.

Product name	Applicable pH range (Normal 20 to 40 °C)	Upper limit temperature	
ProteoSil 200-C18, ProteoSil 100-C18	1-10 *1,2,3	60°C (pH 1-7)	50°C (pH 1-10)
ProteoSil 200-C8	1-10 *1,2,3	60°C (pH 2-7)	50°C (pH 1-9)
ProteoSil HILIC	2-8.5 *2	60°C (pH 2-7)	50°C (pH 2-8.5)
ProteoSil 300-C18 / 300-C8 / 300-C4 / 100-SEC / 300-SEC	2-7.5 *2	60°C (pH 2-7)	50°C (pH 2-7.5)

- \*1 Column life varies greatly depending on conditions such as applicable pH, temperature, and eluent composition. To ensure stable and long-term use, we recommend lowering the column temperature, using low concentrations of buffer salts and additives, and analyzing with eluents containing organic solvents. For analysis at pH 1-2, we recommend using TFA, formic acid, acetic acid, phosphate, etc. For analysis at pH 9-10 within the applicable pH range, we recommend the use of an organic buffer solution of about 5 mM (triethylamine, etc.). Use at pH 2-8 when analyzing only buffer solutions that do not contain organic solvents.
- \*2 In order to prevent premature deterioration, do not to exceed the pH of the eluent described above.
- \*3 For use at pH 1-2 or pH 9-10 within the applicable pH range, we recommend analysis at low temperatures and use of eluents containing organic solvents such as methanol.

### 2-2 UHPLC PEEK / PEEK Column Handling Precautions

- All wetted parts of the UHPLC PEEK / PEEK column are made of resin.
   The column port in particular has a very delicate structure. Therefore, if the column is handled in the same manner as a stainless-steel column, the wetted parts may become damaged.
- Since it can be connected with less torque than a stainless steel column, when connecting to
  equipment piping, etc., make sure that there is no leakage of fluid, and tighten with sufficient torque
  (about 0.8 N-m) to prevent leakage.
- Do not use a male nut with a deformed tip when using it for prolonged periods of time or repeatedly.
   Since the column port may become damaged, we recommend using new resin male nuts when making connections.
- [Recommended fitting: 2-piece type resin male nut.]
- Feeding THF or chloroform through it for a prolonged period of time can lead to premature column degradation.
- When installing or removing the column, use the connecting male nut and column end nut portion.



### 3 Product Features

This product is manufactured under our strict standards at each production process for matrix silica gel, chemical modification, and column performance inspection after packing. This ensures that the product can be used at a consistent level of quality with peace of mind.

### 4 Confirmation of Contents

- Check for any abnormalities in column appearance, packaging, etc.
- · Check for any errors in packing material name, column size, etc
- Check to see that the column performance report is included.
   The column performance report includes information such as the packing material lot No., column serial No., and column performance test eluent. Please keep it in a safe location.
- In principle, columns are sealed with the eluents described in the column performance v
  report.

### 5 Specifications

Туре	Main column	Eluent sealed at shipping
Normal-phase type	ProteoSil 100-SEC / 300-SEC	Hexane / ethanol Mixed eluent
Reversed-phase type	ProteoSil 100-C18 / 200-C18 / 300-C18 / 300-C8 / 300-C4 / 200-C8	Acetonitrile / water Mixed eluent
HILIC type	ProteoSil HILIC	Acetonitrile / water Mixed eluent

### 6 Storage

- If an eluent containing a buffer salt or ion-pair reagent is used in a reversed-phase column, wash it thoroughly with an eluent free of salts.
- Store the reversed-phase column after replacement with an organic solvent such as acetonitrile or methanol.
- When using a reversed-phase eluent, wash ProteoSil 100-SEC or 300-SEC columns with water/acetonitrile in a 50/50 ratio, etc. Store after replacement with 100% acetonitrile.
- Wash the normal-phase column with ethanol or 2-propanol.
   Since alcohol-based solvents are, generally speaking, subject to high pressures during feeding, pay attention to the upper pressure limit of the column and feed it at a reduced flow rate if necessary. Store after replacement with 100% hexane.
- After using a HILIC type column with an eluent containing buffer salts, feed it with an eluent
  with a high water concentration (50% water) to remove hydrophilic substances, and store
  it after replacement with a high acetonitrite concentration (80% or higher acetonitrile).
- When storing the column, tightly plug it with the provided plug and store it in a cool
  place where there is little temperature change or humidity.

\*For UHPLC PEEK / PEEK columns, mount the plug in the same manner as when connecting to equipment, while being careful not to overtighten it.

ProteoSil Series products are manufactured, inspected, packaged, and shipped under strict quality control, but in the unlikely event of a defect, please contact one of our branches, sales offices, or distributors.

However, we do not provide any warranty against damage, problems related to service life, or deterioration caused by use of the product in a manner not in compliance with this instruction manual.

The specification and appearance are subject to change without notice.



#### 高效液相色谱法填充柱

# ProteoSil 系列 使用说明书

### 1 前言

非常感谢您购买本产品。

为使产品充分发挥性能,请仔细阅读并正确使用本使用说明书。

### 2 操作注意事项

### 2-1 使用注意事项

- 请勿掉落或敲击色谱柱。强烈的冲击可能会导致色谱柱劣化。
- 色谱柱由高压匀浆法填充,因此呈现出高耐压性。如果希望长时间稳定使用。
   建议在下表所示的压力范围内使用。

粒径	色谱柱尺寸	推荐压力
1.9 μm, 2 μm	所有尺寸	80 MPa 及以下
3 μm HP	所有尺寸	50 MPa 及以下
3 - 10 μm	内径 0.3 ~ 50 mm	20 MPa 及以下

- 请注意避免压力急剧变化。
  - · 拆下色谱柱时, 请在压力计显示变为 0 之后再拆卸。
  - · 缓慢操作讲样阀时, 会造成色谱柱入口的压力发生急剧变化, 请加以注意。
- 请尽量将样品溶解于与洗脱液组成相同的溶剂(采用梯度方法时为初始溶剂)。
   如果样品溶解在溶解力高于洗脱液的溶剂中,则当大量注入时,分离能力会降低,或者样品会在色谱柱入口处析出。
- 不会相互混合的洗脱液(例:从正相到反相系统)置换时, 请在中间流过异丙醇等中等极性溶剂。此时,醇类溶剂在送液时的压力会变高, 因此请注意色谱柱的上限压力,一边调整流量,一边通过色谱柱容量10倍或以上的液体。
- 若溶出快并出现峰拖尾,其原因可考虑为死体积。 请检查色谱柱接头部分的连接配管是否插入至最深处。 此外,连接至进样器和检测器的配管, 请选择内径与长度均匹配使用色谱柱的内径及其分析系统的配管。 尤其当使用半微型色谱柱等以低流量进行分析时,配管的影响会变大。
- 压力上升和峰裂分的原因可能为色谱柱入口处的过滤器堵塞或脏污。
  - · 请在用不超过 0.45 µm 的膜过滤器等装置过滤洗脱液后再使用。
  - · 请在用不超过 0.45 µm 的针式过滤器等装置过滤样品液后再注入。
  - · 使用滤芯 E 或滤芯 E 2.1x10 mm 超高效液相色谱柱可防止色谱柱堵塞。
- 使用前,请用洗脱液使色谱柱达到充分平衡。
   (使用 HILIC 或离子对试剂的反相模式可能需要一定时间才能稳定。)
- 根据 LC/MS 的分析条件和装置类型等,可能会在色谱柱出口处出现析出物。 使用前请确认析出物的影响。

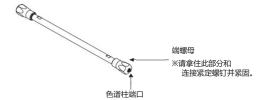
- 色谱柱的 pH 值使用范围、上限温度请参考下表。
   (表内未记载的色谱柱的使用 pH 值范围和上限温度,请参照 GL Sciences 的主页。)
- 超出上限温度使用时,会导致加速劣化,请加以注意。

产品名称	使用 pH 值范围 (常用 20℃-40℃)	上限	温度
ProteoSil 200-C18, ProteoSil 100-C18	1-10 ※1,2,3	60°C (pH 1-7)	50°C (pH 1-10)
ProteoSil 200-C8	1-10 ※1,2,3	60°C (pH 2-7)	50°C (pH 1-9)
ProteoSil HILIC	2-8.5 ※2	60°C (pH 2-7)	50°C (pH 2-8.5)
ProteoSil 300-C18 / 300-C8 / 300-C4 /	2-7.5 %2	60°C (pH 2-7)	50°C (pH 2-7.5)
100-SEC / 300-SEC	2 7.5 M2	00 C (p. 1 Z 1)	50 C (pr. 2 7.5)

- ※1 色谱柱的使用寿命因使用的 pH 值、温度、洗脱液组成等条件而有显著变化。为实现长期稳定使用,建议降低色谱柱温度,使用低浓度缓冲盐和添加剂,用含有机溶剂的洗脱液进行分析。pH1~2 时的分析建议使用 TFA、甲酸、乙酸、磷酸盐等。此外,在 pH 值使用范围内,pH9~10 时的分析建议使用 5 mM 左右的有机缓冲液 (三乙胺等)。仅使用不含有机溶剂的缓冲液进行分析时,请在 pH2~8 的范围内使用。
- ※2 为防止加速劣化,请注意将洗脱液的 pH 值控制在上述范围内。
- ※3 在使用 pH 值范围内以 pH1~2 或 pH9~10 使用时,建议在低温下分析,或者使用含甲醇等有机溶剂的洗脱液。

## 2-2 UHPLC PEEK / PEEK 色谱柱 使用注意事项

- UHPLC PEEK / PEEK 色譜柱接触液体的部分均为树脂材料,这是一个微妙的结构。 色谱柱端口部分的结构非常精密,因此如果按照与不锈钢柱相同的方式处理, 将损坏接液部件。
- 可以用小于不锈钢色谱柱的扭矩进行连接,因此当连接到装置配管等时, 请在确认未发生漏液的同时,以不会发生泄漏的扭矩(标准: 0.8 N·m 左右)紧固。
- 请勿使用因长时间使用或反复使用等而导致前端变形的紧定螺钉。
   色谱柱端口部有破损的可能性,所以建议用新的树脂紧定螺钉进行连接。
   健议装配:两片式树脂紧定螺钉
- 用 THF 或三氯甲烷进行长时间通液时,可能会导致色谱柱加速劣化。
- 安装和拆卸色谱柱时,请拿住连接紧定螺钉和柱端螺母部分。



### 3 产品特点

从母体硅胶、化学修饰到填充后的色谱柱性能检查,本产品在每道工序中均执行本公司独有的 严格标准,质量稳定如一,可以放心使用。

### 4 内容物检查

- 请检查色谱柱的外观、包装等是否有异常。
- 请检查填充剂名称、色谱柱尺寸等是否有误。
- 请确认有随附的色谱柱性能报告。
   色谱柱性能报告,记载有填充剂批次编号、色谱柱序列号、
   色谱柱性能检查洗脱液等,请妥善保管。
- 色谱柱原则上已封入色谱柱性能报告中所记载的洗脱液。

### 5 规格

类型	主要色谱柱	出厂时封入洗脱液
正相系统	ProteoSil 100-SEC / 300-SEC	己烷/乙醇
工怕系列	110 de 310 310 310 310	混合洗脱液
反相系统	ProteoSil 100-C18 / 200-C18 / 300-C18 /	乙腈/水
及怕系统	300-C8 / 300-C4 / 200-C8	混合洗脱液
HILIC 系统	ProteoSil HILIC	乙腈/水
TILIC 新知		混合洗脱液

### 6 保管

- 若在反相色谱柱内使用含缓冲盐或离子对试剂等的洗脱液,请用除去盐的洗脱液充分清洗。
- 反相色谱柱请在用乙腈、甲醇等有机溶剂置换后再保管。
- 若在使用反相洗脱液时需清洗 ProteoSil 100-SEC 或 300-SEC 色谱柱, 请用水/乙腈 = 50/50 的溶液等清洗。

保管时,请在置换成100%乙腈之后再保管。

- 清洗正相色谱柱时,请用乙醇或异丙醇进行通液并清洗。
   一般来说,醇类溶剂在送液时压力会变高,因此请注意色谱柱的上限压力,必要时降低流量后再送液、保管时请在置换为100%已烷之后再保管。
- 以含缓冲盐的洗脱液使用 HILIC 系统色谱柱之后,为去除亲水性物质, 请用水浓度高的洗脱液(水 50%)进行通液,并置换为乙腈浓度高的洗脱液(乙腈 80% 及以上),然后进行保管。
- 保管色谱柱时,请用附带的塞头进行密封,然后保管在温度变化小、湿气少的阴凉处。
   ※若使用 UHPLC PEEK / PEEK 色譜柱,则与连接至装置时相同,需注意不要过紧安装塞头。

ProteoSil 系列均在严格的质量管理下生产、检查、包装和出厂,如发现问题,请联系我们的分公司、营业所或代理店。

不过,有关因破损和使用寿命而引起的问题,以及使用时未按照本使用说明书上的方法而造成的 劣化等,我们无法提供保修。

规格及外观如有变更, 恕不另行通知。



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