

## ARION® COLUMN CARE AND PROTECTION GUIDE

## Introduction

Thank you for purchasing the Arion® HPLC column.

Every ARION® HPLC column is a precision product which will provide excellent performance, reproducibility and column lifetime if cared for properly. The information and recommendations contained in this manual are designed to guide you in the care and use of your column. Please follow the instructions herein to maximize column performance and lifetime. If you have any questions, please contact our technical department.

# Upon receipt of the column

- Verify the column you received is the column you ordered
- Check the column for any physical damage which may have occurred during shipping
- All columns are shipped in the testing solvent, unless otherwise specified Each ARION® HPLC column is individually packed and tested to ensure high column quality. Every column is supplied with its Test Chromatogram. The warranty period is 3 months and begins upon receipt of the column.



## **COLUMN CARE GUIDE**

# **SPECIFICATION**

ARION phases	Particle size (µm)	Por Size (Å)	Surface area (m²/g)	Carbon load (%)	pH stability	Endcapping	100% aqueous mobile phase stability
Plus C18	1.7, 2.2, 3, 5, 10, 15	100	420	18%	1.0 to 10	Multi-step	NO
Polar C18	2.2, 3, 5, 10, 15	120	325	16%	1.5 to 7	Multi-step	YES
C8	3, 5	120	325	11%	2.0 to 7	Single-step	NO
Phenyl-Butyl	2.2, 3, 5	100	300	12%	1.5 to 7.5	Single-step	NO
Biphenyl	3, 5	100	300	12%	1.5 to 7.5	Single-step	YES
PFP	5	100	420	14%	1.5 to 7.5	Single-step	YES
NH <sub>2</sub>	2.2, 3, 5	120	325	5%	2.0 to 6.5	Proprietary	NO
CN	3, 5, 10	120	325	8%	2.0 to 7	Single-step	NO
HILIC Plus	2.2, 3, 5	100	420	-	1. to 7	Proprietary	NO
Si	2.2, 3, 5, 10	100	420	-	1.5 to 7	-	NO
SAX	5	120	325	-	1.0 to 7.5	-	YES
SCX	5	120	325	-	1.0 to 7.5	-	YES



## Mobile phase consideration

- The correct direction of the solvent flow is indicated by an arrow on the column identification label.
- Use only HPLC or LC/MS grade solvents and water
- Use only highest purity chemicals and reagents
- Filter and degas all mobile phases and sample prior to use through a suitable membrane filter
- Repetitive replacement among solvents with a large difference in polarities might degrade the column performance
- In general, organic solvents like acetonitrile, methanol and tetrahydrofuran (THF) are recommended for regular use. When using THF as a mobile phase, be mindful of the solvent resistance of your system or tubing (PEEK parts are especially unsuitable for use with THF).
- Recommendations of pH stability of the column are shown in the specification table. When using the column at pH near the upper or lower limit, column lifetime may be shortened under certain conditions by temperature and mobile phase composition.

# Shipping solvent and column storage

- The shipping solvent is specified on the "Certificate of Analysis", which is enclosed with each column
- Make sure solvents are miscible using solvents that are immiscible with the solvent in the column can permanently damage the column.
- Salt and buffer precipitation from the mobile phase can permanently damage the column.
- Never store columns containing buffers or ion-pairing reagents.
- Flush with 10 column volumes of mobile phase without buffer to remove any buffers or salts
- Use storage solvent:

70% Acetonitrile / 30% Water for C18, C8, Biphenyl, PFP, Phenyl column

stationary phase

Ethanol for Normal Phase Silica, NH<sub>2</sub>, CN, SAX, SCX

column stationary phase

70% Acetonitrile / 30% Water for HILIC column stationary phase

## Column cleaning

Arion® HPLC/UHPLC columns mostly use different frits on the stainless-steel tube end, which means that the **reversed flow to clean the columns is NOT recommended.** 

Before starting any kind of cleaning procedure, make sure your in-column solvent or mobile phase is miscible with the recommended cleaning solvent. Flow rates should be 1/5 - 1/2 of the typical flow rate.

### Method for reversed phase (C18, C8, Phenyl, Biphenyl, PFP, CN, NH<sub>2</sub>)

Rinse with 10 Column Volumes each of:

- 95 % Water/5 % Acetonitrile (for buffer removal)
- IPA (THF)
- 95 % Acetonitrile/5 % Water
- Mobile Phase

A cleaning solution containing THF might be effective when removing highly hydrophobic (lipid soluble) substances. Be mindful of the solvent resistance of your system or tubing, PEEK parts are especially unsuitable for longer use with THF.

## Method for bonded normal phase (CN, NH<sub>2</sub>)

Rinse with 10 Column Volumes each of:

- Chloroform
- Isopropanol
- Methylene Chloride
- Mobile Phase

### Method for unbonded silica (Si)

Rinse with 10 Column Volumes each of:

- Hexane
- Methylene Chloride
- Isopropanol
- Methylene Chloride
- Mobile Phase

Water removal procedure:

Flush column with 30 mL 2.5 % 2,2-dimethoxy-propane and 2.5 % glacial acetic acid in Hexane

#### **Method for HILIC**

Rinse with 10 Column Volumes each of:

- 95 % Water/5 % Acetonitrile (for buffer removal)
- 95 % 100 mM Ammonium Acetate, pH 5.8/5 % Acetonitrile
- 95 % Water/5 % Acetonitrile
- Mobile Phase

### Method for SO<sub>3</sub>H bonding phase (SCX)

To break ionic interactions, the pH has to be at 2 (partial protonization of SCX) or at 8 (sometime not sufficient for basic compounds with pka >9.5). The pH stability of SCX silica is between 1.0 to 7.5, this is limiting the efficiency of the washing. Washing procedure:

- Washing 15-20 column volumes at pH 2.5 with formic acid + water + 30% organic solvent (Target: remove buffer + eliminate a part of weak basic compounds and sometime a part of stronger basic compounds + eliminate non ionic compound)
- Washing 15 column volumes at pH 8 with volatile additive (no salt buffer) + water
- + 30% organic solvent (Target: eliminate another part of weak basic compounds + eliminate non ionic compounds)
- Washing 7 column volumes with 50/50 Water/organic solvent without additive + 70/30 Water/organic solvent (Target: column ready to be store)

## Method for quaternary amine bonding phase (SAX)

To break ionic interactions, the pH has to be at 2 (50% of form for compounds with pka at 3) or at 8 (not sufficient for SAX bonding). The pH stability of SAX silica is between 1.0 to 7.5, this is limiting the efficiency of the washing. Washing procedure:

- Washing 15-20 column volumes at pH 2.5 with formic acid + water + 30% organic solvent (Target: remove buffer + eliminate a part of weak acid compounds and sometime a part of stronger acidic compounds + eliminate non-ionic compound)
- Washing 15 column volumes with 50/50 Water/organic solvent without additive
- + 70/30 Water/organic solvent (Target: column ready to be store)



# Column Protection = ARION® Guard System (AGS)

The ARION® Guard System (AGS) is a universal guard system, which can be connected to almost any column hardware on the market. It is easy to use and it offers the shortest retention time shift of analytes in comparison with other major manufacturers. The AGS consists of a guard holder and guard cartridge, which is offered with various silica materials according to the stationary phase in the HPLC column used.



## How to tighten AGS to column

As the AGS is not Finger-tight, it is necessary to tighten the holder with a wrench. First, tighten the AGS housing with wrench until it is snug (it is possible to use wrench No. 11, or 7/16 can also be used) followed by approx. 90° turn in order to seal it.

# How to tighten AGS housing

Maximum torque to tighten the AGS holder (mail housing to female housing) is approximately 6.5 to 8 Nm.

# Port compatibility

This holder works for Agilent, Halo, Phenomenex, Parker, Chromservis and most other manufacturers except Waters.



### Main features and benefits

- Universal fits virtually any column
- The lowest influence on retention times compared with other guard systems
- Small size for easier installation in the column oven
- Any orientation of the cartridge
- Pressure rating up to **900 bars** (holder)

Maximum pressure of PEEK ferrules depends on the tubing used. Maximum pressure of **400 bar** is for 1/16 OD tubing with ID 0.05 to 0.18 mm. ARION® Guard System with Stainless Steel ferrule can be used up to **689 bars**.



### Other environments

- The operating pressure should be kept under **350 bar** for fully porous particles **3.0 and 5.0 \mu m**.
- 2.2 µm particles have a pressure resistance of up to 700 bars
- 1.7 µm particles have a pressure resistance of up to 1000 bars
- Avoid using a column repeatedly near the pressure limit or abrupt changes in pressure to prevent shortening of the column life.
- We recommend using a pre-column filter to prevent the column frit from being clogged with samples.
- Suggested maximum temperature for Arion® LC columns is 100°C, however temperature limits are dependent on your running parameters.

# Ordering information

Complete ordering information is available at <u>www.chromservis.eu</u> or <u>www.arionchromatography.com</u>.