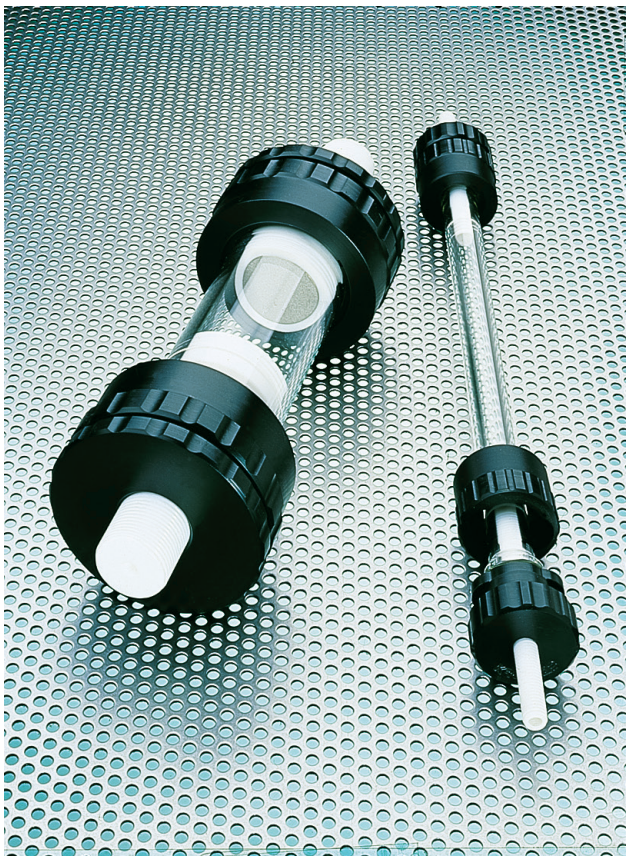
YMC EUROPE GMBH

Manual

Medium Pressure Chromatography Columns

ECO^{PLUS} Series



These instructions should be read prior to using the column!

Also available to download at: *http://ymc.de/ecoplus-glass-columns.html*

Operating instructions

This manual ensures the safe and efficient use of the medium pressure ECO^{PLUS} series of chromatography columns. The manual is part of the product and is available to download at *http://ymc.de/ecoplus-glass-columns.html*.

The user must read this manual carefully and have fully understood its content before attempting to use the column. A prerequisite for the safe operation is the compliance with all the specified safety and operating instructions in this manual. The safety instructions are described in chapter 1.

In addition, all the local occupational health and safety and general safety regulations applying to the use of the product must be complied with.

The following laws and standards were taken into account in the creation of the manual:

- 2001/95/EG
- DIN EN 82 079-1
- VDI 4500 Section 1

The terms and conditions of YMC Europe GmbH apply.

Copyright protection

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1. Safety

This section gives an overview of all important safety aspects for the protection of users and the safe and trouble-free operation of ECO^{PLUS} columns. Other task-specific safety requirements are included in the sections for the specific operations. In addition, please also comply with your national legislation and other applicable safety regulations.

1.1. Hazard Symbols

Hazard symbols are highlighted by a combination of symbols and keywords. For your safety, always read the complete text of the symbol.

Safety information is given in these instructions by use of a combination of symbols and key words that describe the nature and severity of the hazard:

⚠ DANGER

This combination of symbol and key words indicates a potentially hazardous situation that may result in death or serious injury if ignored.

⚠ WARNING

This combination of symbol and key words indicates a potentially hazardous situation that may result in serious or fatal injury if ignored.

⚠ CAUTION

This combination of symbol and key words indicates a potentially hazardous situation that may result in minor or moderate injury if ignored.

ATTENTION

Failure to observe this warning may result in property damage.

All symbols are constructed in the same way. They include the signal word, type and source of danger, results of ignoring them as well as actions to prevent or avoid the danger.

1.2. Composition of operating instructions

Operating instructions require specific actions. Always follow each single action step in the prescribed order.

Operating instructions are designed in the following way and are characterised with the relevant symbols:

► Aim of the operating instruction

1. Action step

   ✓ Impact of the action step to control if the step was performed correctly

2. Next action step

☑ Result of the whole operation instruction

1.3. Areas of application

By careful selection of high-quality inert materials, ECO^{PLUS} glass columns can be used in both the normal phase and reversed phased modes, for analytical and preparative scale. ECO^{PLUS} glass columns are frequently used for biochromatography separations with the appropriate stationary phases. They allow high recovery of the analytes due to their low interaction with such biomolecules (e.g. proteins, nucleic acids or oligosaccharides). The columns are easy to operate due to the “Quick-Lock” closures and have very adjustable volumes due to the two adjustable plungers.

With a wide range of diameters, volumes, lengths and frit porosities, a wide variety of applications can be performed with ECO^{PLUS} glass columns.

ECO^{PLUS} glass columns meet the highest criteria for professional use in the laboratory.

Special emphasis has been placed on the wide range of volumes possible (0.6 to 980 ml) and high pressure resistance (up to 80 bar) , so that high flow rates and throughput / efficiency can be achieved.

1.4. Intended Use

The ECO^{PLUS} series of medium pressure chromatography columns is designed for the chromatographic separation of compounds. Their routine use requires no tools. Their correct use requires compliance with all information contained in this guide. If used outside the scope of this intended purpose, it will be considered as misuse.

1.5. Operator requirements

The various tasks described in this manual make different demands of the people who carry them out and require different levels of training.

⚠ WARNING

Insufficiently trained people may cause dangerous situations.

People with insufficient training cannot accurately estimate the dangers of using the product and therefore could expose themselves and others to the risk of serious injuries.

Technically ECO^{PLUS} glass columns technically are pressure vessels with variable volume. The user must therefore be trained in dealing with pressure equipment. Also, the glass columns are used essentially for the purification of chemical substances. Therefore, the user must also be laboratory trained. Every user must have read and understood this manual before using the glass columns.

1.6. Personal protective equipment

Personal protection equipment serves to protect people from health and safety risks at work. Staff must wear personal protective equipment during work on and with the column according to national and internal laboratory safety regulations required for such work, for example safety goggles and lab coats.

2. Delivery

2.1. Quantities delivered

Fig. 2.1: A column in its packaging as delivered

Please take the complete glass column from the packaging. Compare the column label on the glass body with the details in the delivery note (part number (Pr.-No.), inner diameter (ID), maximum bed length (Length), maximum pressure (Pressure) and serial number (Ser.-No.)). This information must match. If this is not the case, please immediately contact YMC Europe GmbH.

YMC EUROPE GMBH	D-46539 Dinslaken
Pr.-No.:	TAC25/250LS0-SR-2
ID/Length:	25 x 250 mm
Pressure:	50 bar
Ser.-No.:	TAC123456

Fig. 2.2: Example of a column label

The ECO^{PLUS} glass columns are transported as pre-assembled units. Each ECO^{PLUS} column is supplied with the appropriate accessories (see table 2, p. 10-11), a certificate of pressure resistance and information material (quick guide).

Based on the column label on the glass body, the actual version can be identified so that you will receive individual support for a special column, if necessary.

The part number (Pr.-No.) of the column contains information about the inner diameter, the maximum length of the bed, the plunger type, the frit material, the porosity of the frit and the column type. The part number is generated from these parameters. An overview of this information, as well as an example of how to interpret the exact type of column, is shown on page 9. This example gives the information for a solvent resistant ECO^{PLUS} column with an inner diameter of 25 mm, a maximum bed length of 500 mm, 2 short plungers and stainless steel frits with a porosity of 2 microns. Moreover, the column is a solvent resistant version.

Table 1: column versions and part numbers

Combination possibilities	**TAC05/** (5 mm ID) **TAC10/** (10 mm ID) **TAC15/** (15 mm ID) **TAC25/** (25 mm ID) **TAC35/** (35 mm ID) **TAC50/** (50 mm ID)	**125** (125 mm max. bed length) **250** (250 mm max. bed length) **500** (500 mm max. bed length)	**—** (Standard-Version has 2 short plungers - no letter is inserted) **SL** (1 short/1 long plunger) **L** (2 long plungers)	**PE** Polyethylene (AB-Version) **G** Sintered glass (SR-Version with ≤ 15 mm ID) **S** Stainless steel (SS) (SR-Version with ≥ 25 mm ID)	**2** (2 µm) **5** (5 µm, only Poly-ethylene Frits) **0** (10 µm)	**-AB-2** (aqueous buffer) **-SR-2** (solvent resistant)
Example Pr.-No. (part number)	**TAC25/**	**500**		**S**	**2**	**-SR-2**
ID	25 mm					
Max. bed length		500 mm				
Type of plunger			Standard version (2 short plungers)			
Frit material				Stainless steel		
Frit porosity					2 µm	
Version						SR version

On request, special versions of the columns can be manufactured. YMC Europe GmbH would be happy to discuss your requirements.

2.2. Delivery inspection

Delivery check

▶ This is how you proceed during delivery inspection:

1. Compare the column label on the glass body with the details on the delivery note.
2. Check the completeness of the delivery using the „Checklist Scope of Supply“ ▶*Chapter 2.1, p. 8.*
3. Check the delivery for externally identifiable transport damage.

Transportation damage

▶ In case of visible damage, proceed as follows:

1. Do not accept the delivery or accept only conditionally.
2. Record the extent of damage on the transport documents or on the delivery note of the carrier.
3. Inform YMC Europe GmbH and/or the responsible customer consultant about the damage.

If any transportation damage is noted, claim as soon as it is detected.

We recommend that you keep and use the original packaging and the accompanying foam pads (e.g. polystyrene, foam, etc.) for storage to ensure safe transport of the column is possible.

Table 2: Check list for quantity of parts delivered

	Delivered	Number	Description
For ID 5 mm columns		1 m	PT0.8FE1.6 1/16 inch FEP tubing (0.8 x 1.6 mm, assembled with sleeve support)
		4	JR-55050 1/4 inch - 28 fixing screw for 1/16 inch ferrule For connection to the column
		4	JR-CFL-CB1KF-S Ferrule for 1/16 inch tubing + JR-55050
		2	K.P207 M6 fixing screw for 1/16 inch tubing For the connection to a GE-ÄKTA-System
		2	K.P200N Ferrule for 1/16 inch tubing + K.P207
		2	JR-58000 Fixing screw 10-32 thread
		2	K.P316 plug for 1/4 inch - 28G fitting
		1	Quick guide
		1	Certificate of pressure resistance

Table 2: Check list for quantity of parts delivered

	Delivered	Number	Description
For ID 10-15 mm columns		1 m	PT0.8FE1.6 1/16 inch FEP tubing (0.8 x 1.6 mm, 40700)
		4	JR-55050 1/4 inch - 28 G fixing screw for 1/16 inch ferrule For connection to the column
		4	JR-CFL-CB1KF-S Ferrule for 1/16 inch tubing + JR-55050
		2	K.P207 M6 fixing screw for 1/16 inch tubing For connection to a GE-ÄKTA system
		2	K.P200N Ferrule for 1/16 inch tubing + K.P207
		2	JR-58000 Fixing screw 10-32 thread
		1	Frit removal tool
		2	K.P316 Plug for 1/4 inch - 28G fitting
		1	Quick guide
		1	Certificate of pressure resistance

Table 2: Check list for quantity of parts delivered

	Delivered	Number	Description
For ID 25-50 mm columns		1 m	PT1.6FE3.2 1/8 inch FEP tubing (1.6 x 3.2 mm)
		4	JR-55051 1/4 inch-28 G fixing screw for 1/8 inch ferrule For connection to the column
		4	JR-CFL-CB2KF-S Ferrule for 1/8 inch tubing + JR-55051
		2	K.P307 M6 fixing screw for 1/8 inch ferrule For connection to a GE-ÄKTA system
		2	K.P300N Ferrule for 1/8 inch tubing
		1	Frit removal tool
		2	K.P316 Plug for 1/4 inch - 28G fitting
		1	Quick guide
		1	Certificate of pressure resistance

3. Start-up

3.1. Precautions for use

⚠ WARNING

Use the appropriate protective clothing when working with the glass column, to ensure the safety of the user, e.g. safety glasses and lab coat.

⚠ WARNING

The column must never be used under gas pressure.

⚠ CAUTION

Only operate the column at pressures corresponding to the published specifications. Otherwise there is a risk of material damage and even injury to the user.

⚠ CAUTION

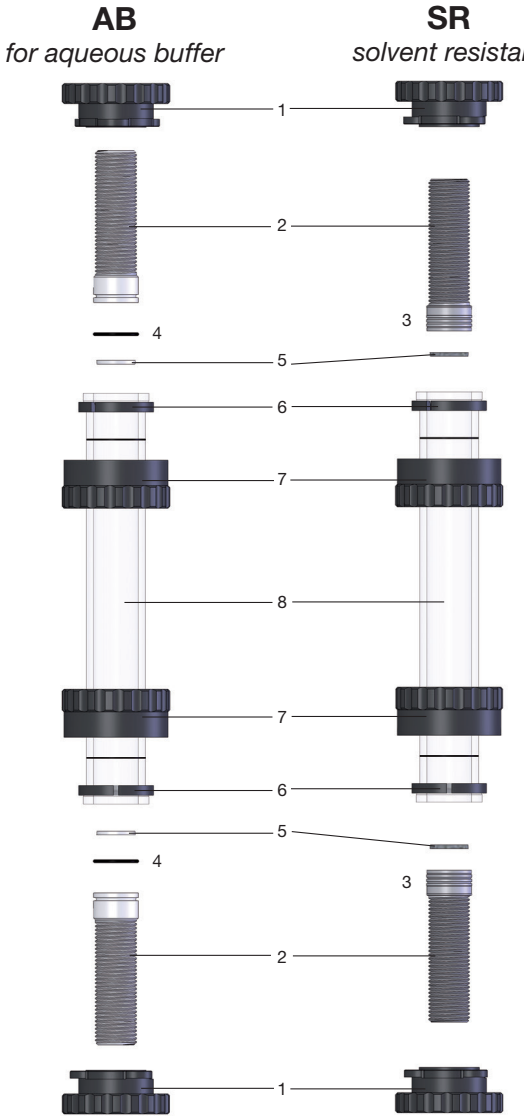
The temperature range used must not exceed the specified limits. (▶*Chapter 3.3, Specifications*).

⚠ CAUTION

Take care when working in the immediate vicinity of the column.

Do not use column with chemicals that are not suitable for use with the wetted materials. (see table 5, p. 23).

3.2. Column construction



1 counter screw (bayonet fixing)
2 plunger
3 lamellar seal
4 O-Ring
5 frit
6 retaining ring
7 counter nut (bayonet fixing)
8 glass body

Fig. 3.1. Detailed drawing of ECO^{PLUS} column. Left - aqueous buffer column version (Version AB), Right - solvent resistant column version (version SR).

The ECO^{PLUS} chromatographic column consists of a glass body (8), into which plungers are attached at both ends (2). These plungers are attached to a lock screw (1) and lock nut (7) held in place by the snap rings (6). The frits (5) are inserted in the end recesses of the plungers.

The ECO^{PLUS} columns are available in two versions. The Solvent Resistant (SR) version is equipped with an innovative Teflon lamellar sealing system and can be used with all common organic solvents. The Aqueous Buffer (AB) version is designed for use with aqueous buffer systems in chromatography and uses a suitable O-ring seal.

3.3. Specifications

Aqueous buffer (AB) version

Temperature range	4 - 40 °C
Plunger	PE (polyethylene)
Seals	O-ring, EPDM (ethylene propylene diene monomer rubber)
Frits	PE (polyethylene)
Height adjustment	2 short plungers, 1 short and 1 long plunger, 2 long plungers (see table 3)
Connection for tubing	1/4 inch-28G female screw thread
Tubing	FEP tubing

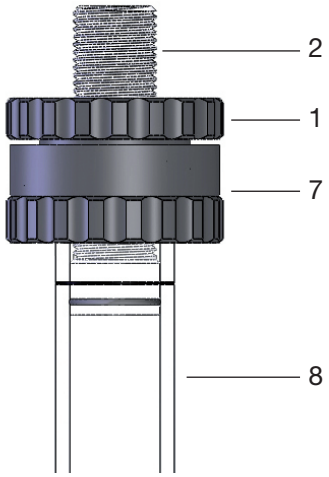
Solvent resistant (SR) version

Temperature range	16 - 40 °C
Plunger	PTFE (polytetrafluoroethylene)
Seals	ID 5 - 35 mm: PTFE triple chevron lamellar seals
	ID 50 mm: O-ring, Kalrez® (colour: white)
Frits	ID 5 - 15 mm: sintered glass
	ID 25 - 50 mm: stainless steel
Height adjustment	2 short plungers, 1 short and 1 long plunger, 2 long plungers (see table 3)
Connection for tubing	1/4 inch-28G female screw thread
Tubing	FEP tubing

Table 3: Specifications of pressure, bed length, volume

ID [mm]	Pressure limit [bar]	2 Short plungers		1 Short and 1 long plunger		2 Long plungers	
		Bed length [mm]	Volume [ml]	Bed length [mm]	Volume [ml]	Bed length [mm]	Volume [ml]
5	AB = 80 SR = 80	30 - 125 150 - 250 400 - 500	0.6 - 2.4 3.0 - 4.9 7.9 - 9.8	0 - 125 70 - 250 320 - 500	0 - 2.4 1.4 - 4.9 6.3 - 9.8	0 - 125 0 - 250 240 - 500	0 - 2.4 0 - 4.9 4.8 - 9.8
10	AB = 80 SR = 50	40 - 125 160 - 250 410 - 500	3.2 - 9.8 13 - 19 33 - 39	0 - 125 80 - 250 330 - 500	0 - 9.8 6.2 - 19 26 - 39	0 - 125 0 - 250 250 - 500	0 - 9.8 0 - 19 20 - 39
15	AB = 70 SR = 50	30 - 125 155 - 250 405 - 500	6 - 22 28 - 44 72 - 88	0 - 125 75 - 250 325 - 500	0 - 22 14 - 44 58 - 88	0 - 125 0 - 250 245 - 500	0 - 22 0 - 44 44 - 88
25	AB = 50 SR = 50	30 - 125 160 - 250 410 - 500	15 - 60 80 - 120 205 - 245	0 - 125 80 - 250 330 - 500	0 - 60 40 - 120 165 - 245	0 - 125 0 - 250 250 - 500	0 - 60 0 - 120 125 - 245
35	AB = 40 SR = 40	35 - 125 160 - 250 410 - 500	35 - 120 155 - 240 395 - 480	0 - 125 80 - 250 330 - 500	0 - 120 80 - 240 320 - 480	0 - 125 0 - 250 250 - 500	0 - 120 0 - 240 245 - 480
50	AB = 30 SR = 15	40 - 125 170 - 250 415 - 500	80 - 245 335 - 490 815 - 980	0 - 125 90 - 250 340 - 500	0 - 245 180 - 490 670 - 980	0 - 125 0 - 250 255 - 500	0 - 245 0 - 490 500 - 980

3.4. Dismantling



▶ Procedure for dismantling:

1. With the column initially NOT open.
2. Turn the counter screw (1) one-half turn clockwise, so that the plunger moves inside the column away from the packed bed.
3. Only now open the bayonet fixing. Press counter screw (1) against counter nut (7) and turn the screw a quarter-turn counter-clockwise. Then disconnect the counter nut (7) from the counter screw (1).
4. Remove the plunger together with the counter screw (1) from the column body (8).

If the plunger is still too tightly fitted in the glass body (8), it can be extracted by using the counter screw to loosen the seal.

5. Remove the retaining rings from the glass body ends.
6. The counter nuts (7) can now also be removed from the glass body (8).

3.5. Cleaning

▶ When cleaning the ECO^{PLUS} column please follow these steps in sequence:

1. Dismantle the column (broken down into column tube (8), plungers (2), counter screws (1) and counter nuts (7))
2. Clean column tube and plungers in an ultrasonic bath using doubly distilled water. After use, please wash the column body and plungers in 0.5 M sodium hydroxide solution or 0.1 - 0.5% sodium hypochlorite solution (in water, v/v).
3. Clean the plunger frits in the plunger by flushing with 0.1 - 0.5% sodium hypochlorite or rinse with 0.5 M sodium hydroxide solution for 30 - to 60 minutes.

We recommend you record when the cleaning procedure has been performed. For this purpose, we have provided you with the logbook in the Appendix. More documents for download at http://ymc.de/ ecoplus-glass-columns.html.

3.6. Assembly

Before this step, please first take the column apart and clean its component parts thoroughly (▶Chapter 3.4, Dismantling and ▶Chapter 3.5, Cleaning).

ATTENTION

Please do not use any parts with the ECO^{PLUS} column that were not provided by YMC (▶Chapter 2.1, quantities delivered), or are not listed in the spare parts list (▶Chapter 9.1, spare parts lists). Otherwise there is a risk of damage to the column. Only when using the specified parts the function is assured.

3.6.1. Installing the plunger

Before inserting the plunger into the glass tube, clean the seals and wet them with the eluent to be used.

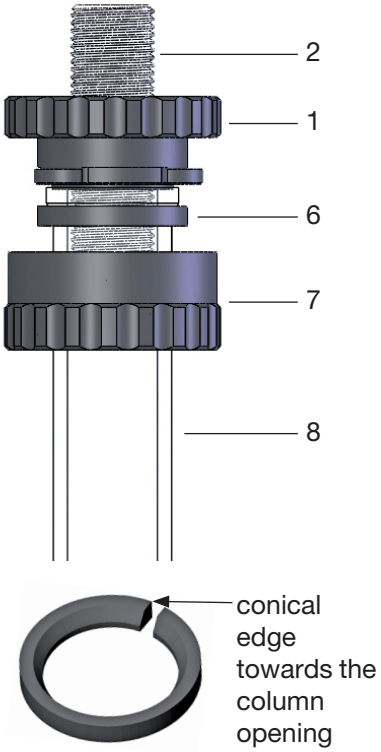
▶ The plungers are inserted in the following sequence:

1. The counter nut (7) is put over the glass body (8) and the retaining ring (6) is put in place with its conical edge toward the column opening.

ATTENTION

Danger of damage! When inserting the plunger, it is important to ensure that the plunger is vertical and not tilted as this could affect its sealing ability.

2. Introduce the plunger (2) with the counter screw (1) attached carefully into the glass tube without tilting.
3. Align the counter screw (1) and counter nut (7) by rotating them until the stud on the bayonet counter screw aligns with its receiving slots in the counter nut (7).



ATTENTION

The column must be operated only with securely locked end pieces. Otherwise, the plunger can be forced out of the column due to the pressure inside.

4. Close the column with a quarter clockwise turn of the counter nut (7) relative to the counter screw (1). The bayonet fitting locks into place with an audible click.

ATTENTION

Turning of the counter nut (7) can lead to opening of the bayonet. To adjust the height of the plunger, only turn the counter screw (1)!

3.6.2. Column to tubing connections

ATTENTION

Only the 1/4 inch-28G fixing screws should be used for the connection of tubing to ECO^{PLUS} glass columns! Use of other screw threads will cause damage to the threads in the plungers.

ATTENTION

Ensure that the appropriate tubing is used. The internal diameter must be suitable for the desired flow rate and the tubing resistant to the solvent used. The use of unsuitable tubing may cause compression of the column bed (dead volume). Please check the compatibility of the tubing used before connecting the column to the chromatography system or pump with the fixing screws.

ATTENTION

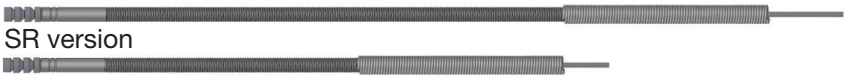
Always cut the tubing with the tube cutter *(▶Chapter 9.1, parts lists)* and never use scissors, as this will squash the tubing. It is important that the tubing is cut at 90°, since the cut surface is an integral part of the sealing surface.

For connection of tubing to the column 1/4 inch-28G screws are used. 4 pieces are supplied. Depending on the internal diameter of the column the appropriate fixing screws are supplied. Screw fittings with other types of threads are not suitable for connection to the glass column and will damage the threads in the plungers. The following fixing screws are supplied for the different versions.

TAC05

Connection of tubing to an ECO^{PLUS} column with an internal diameter of 5 mm (TAC05) is not necessary, because these columns are supplied with pre-fitted tubing.

AB version

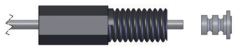


SR version

TAC10/TAC15

For tubing with 1.6 mm outer diameter

JR-55050	Fixing screw 1/4 inch-28 G (4 pieces in accessory pack.)
JR-CFL-CB1KF-S	Collapsible ferrule (4 pieces in accessory pack.)



TAC25/TAC35/TAC50

For tubing with 3.2 mm outer diameter

JR-55051	Fixing screw 1/4 inch-28 G (4 pieces in accessory pack.)
JR-CFL-CB2KF-S	Collapsible ferrule (4 pieces in accessory pack.)

▶ When assembling the fittings please follow these instructions:

1. Cut tubing at 90° with a tubing cutter (e.g. Clean-Cut tubing cutter, ▶Chapter 9.1, Spare Parts Lists).
2. Insert the tubing through fixing screw (see above).
3. Insert the ferrule with the tapered side against the fixing screw onto the tubing (see above). If the ferrule does not fit on the tubing gently expand the tapered end with a suitable pointed tool (such as a tack or nail).

ATTENTION

Be careful to make sure that the fixing screws are straight and properly seated when screwing them into the plunger. Otherwise you will destroy the female threads of the plunger.

4. Insert the fitting into the plunger. Tighten the screw until pressure is felt, then tighten further approximately half a turn.

3.6.3. Connection to LC Systems

ATTENTION

Please observe the notes on tubing handling from the previous section to connect the column to the system!

The accessories supplied include additional fixing screws. These include two each 1/4 inch-28G, M6 and 10-32 fixing screws and corresponding ferrules. *(▶Chapter 2.1, p. 10-11)*. These fixing screws can be used to connect the tubing attached to the column to a chromatographic system. Please check the technical specifications of the system used. According to these technical specifications, you can use the appropriate fixing screws from the YMC accessory pack.

3.7. Pressure test

This test procedure should be used to check the tightness of both the seals of the ECO^{PLUS} column and also the tubing connections. Before using the column for chromatographic applications, it is recommended that a pressure test is performed to demonstrate correct assembly. Prior to delivery of the column, this test has been carried out according to the standard procedure listed below. The corresponding certificate has been included.

⚠ WARNING

The glass column must never be used with gas pressure! There is a risk of damage to the column or may result in serious injury if ignored! Please comply with the specified pressure limits of columns and accessories.

ATTENTION

Before operation, please make sure that the bayonet locks are fully closed! If this is not the case, there is a danger that the plungers will be forced out of the column. Therefore, always check that the bayonet locks are engaged.

Standard operating procedure

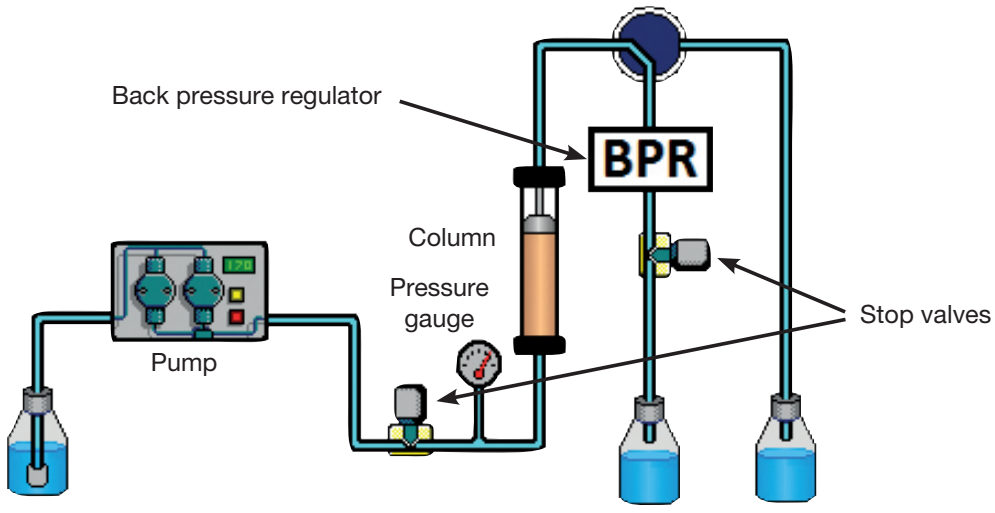


Fig. 3.2: Schematic overview of a system for a pressure test

▶ This is how to perform the pressure test:

1. Take the appropriate safety measures to protect the user in the event that the test fails.
2. Place the column and suitable pump in a closed cabinet (fume hood) and attach a pressure gauge before the column (Fig. 3.2).
3. Fill the column with isopropanol or water.
   The AB version of ECO^{PLUS} columns is inert to isopropanol, so that the pressure test can also be performed safely on the AB version with isopropanol.

ATTENTION

Check on the label and / or the delivery documents for the maximum allowable pressure of the column. Exceeding the maximum pressure can result in damage to the column body and plungers.

4. Now increase the pressure in the column up to the maximum permissible value by slow, gradual adjustment of the flow rate of the pump. Close the stop valves in order to maintain the appropriate maximum pressure.
5. Hold the maximum pressure constant for 10 min. This is to test for leaks or damage.
   Document the test with respect to the duration and the initial and final pressures.

If leaks arise, please check the connections and replace tubing, fittings or locking screws.

Leaking seals are usually due to particulate contamination between the seals and the column walls!

3.8. Chemical resistance

The chemical resistance is dependent on the wetted materials. The chemical stability of the column can be worked out from the chemical resistance of the respective materials used. These materials can be found by correlating the ending of the column part number with tables 4 and 5.

ATTENTION

The AB version has only limited resistance to organic solvents. Before you use organic solvents with ECO^{PLUS} AB version columns, check the resistance of the materials used against the corresponding solvent.

Table 4: Used materials (x=frit porosity)

	Ending of part number	Materials used						
		PE	PTFE	Viton®	EPDM	FEP	Sintered glass	Stainless steel
Possible combinations	PEX-AB-2 (aqueous buffer)	✓		Optional	✓	✓		
	GX-SR-2 (SR.Version with ≤ 15 mm ID) (solvent resistant)		✓			✓	✓	
	SX-SR-2 (SR-Version with ≥ 25 mm ID) (solvent resistant)		✓			✓		✓

ATTENTION

In solvent mixtures, the resistance may differ considerably from those of the pure solvent.

ATTENTION

Such statements are based on our current technical knowledge and experience, as well as on the existing literature and information provided by the raw material manufacturer. However, these do not free the user of our products from their own testing. Certain properties or uses may not be derived either directly or implied from our information.

Table 5: Chemical resistance of materials at 20°C

Solvent	PE	PTFE	Viton®	Kalrez®	EPDM	FEP	Sintered glas	Stain-less steel
Acetone	✓	✓	✗	✓	✓	✓	✓	✓
Acetonitrile	✓	✓	○	✓	○	✓	✓	✓
Ammonium dihydrogen phosphate	✓	✓	○	✓	✓	✓	✓	✓
Cyclohexane	✓	✓	✓	✓	✗	✓	✓	✓
Dichlorme-thane	✗	✓	○	✓	✗	✓	✓	✓
EDTA	✓	✓	✓	✓	✓	✓	✓	✓
Acetic acid (5%)	✓	✓	○	✓	✓	✓	✓	✓ L
Ethanol	✓	✓	✓	✓	✓	✓	✓	✓
Ethyl acetate	✓	✓	✗	✓	○	✓	✓	✓
n-Hexane	✓	✓	✓	✓	✗	✓	✓	✓
Isopropanol	✓	✓	✓	✓	✓	✓	✓	✓
Methanol	✓	✓	○	✓	✓	✓	✓	✓
Sodium hydroxide (45%)	✓	✓	○	✓	✓	✓	✓	✓ L
1 M hydrochloric acid	✓	✓	✓	✓	✓	✓	✓	✗

✓ = stable ○ = limited ✗ = unstable L = crevice corrosion possible

If the desired solvent is not listed, please contact YMC for assistance.

Contact information:
YMC Europe GmbH, Schöttmannshof 19, D-46539 Dinslaken,
Phone: +49 (0)2064 427-0, Fax: +49 (0)2064 427-222, E-Mail: info@ymc.de, www.ymc.de

4. Operation

Chromatography columns can be packed by various methods. This is dependent on column packing material and solvents used.

Two different methods can be distinguished. The so-called sedimentation method, in which the column material is poured into the column as a slurry, which is then allowed to settle under gravity (sedimentation).

The other method, referred to as slurry packing, uses flow under pressure with solvent. The slurry packing method is described as a general recommendation. Both methods are described below.

4.1. Notes on operation

⚠ WARNING

The glass column must never be used under gas pressure! There is a risk of damage to the column or may result in serious injury if ignored! Please do not exceed the recommended pressure limits of columns and accessories.

⚠ CAUTION

Particles in the solvent can lead to clogging of the frits or destruction of the column packing.

- **Use only degassed and pre-filtered solvent.**
- **Ensure that the particle size of the chromatography media used is significantly larger than the pore size of the frit used.**

ATTENTION

Adjust the packing procedure from the information from manufacturer of the packing material. Using incorrect packing conditions can damage or completely destroy the materials.

ATTENTION

Depending on the nature of the packing material a magnetic stirrer can cause mechanical damage to the material. Please contact the manufacturer of the packing material.

ATTENTION

Always use the column within the permissible pressure range! The pressure limits can be found on the delivery note and on the column label. Exceeding the limit may damage the column.

ATTENTION

Use the column only within the permitted working temperature range to ensure the sealing ability.

Operating temperature range:

- **SR version (lamellar seals): 16 - 40 °C**
- **AB version (O-rings): 4-40 °C**

(▶*Chapter 3.3, p. 14*).

ATTENTION

Before operation, please make sure that the bayonet locks are fully closed! If this is not the case, there is a danger that the plungers will be forced out of the column. Therefore, always check that the bayonet locks are engaged.
(▶*Chapter 3.7, p. 20*).

ATTENTION

Only use chemicals in the column, which the column is resistant to. (▶*Chapter 3.8, p. 23*).

ATTENTION

Take care to adjust the height of the plunger with smooth turning of the lock nut and screw, otherwise the ”Quick-Lock” fitting may be inadvertently opened.

4.2. Packing operations

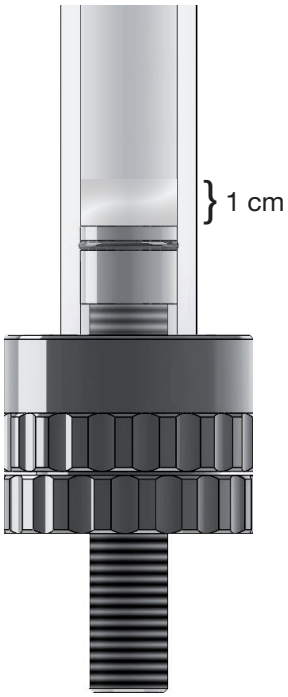
When packing please follow the numbered steps below:

If desired, the packing adapter (available separately) should be assembled. More information about the packing adapter can be found in chapter 4.3 (Packing adapter). Next, the slurry for the final column packing is prepared. Packing of the column may be carried out by various methods. Two procedures are described in this manual. After packing the column, a column test should be carried out to determine performance criteria ▶*Chapter 4.2.4., p. 31*.

4.2.1. Preparing the column

▶ Preparing the column:

1. The column outlet is connected to the pump and an appropriate solvent (or buffer) is added to a depth of 1 cm with appropriate solvents or buffer is present. This ensures that there is no air trapped under or within the frit.
2. The column outlet is sealed with the plug supplied.



Optional: Assembly of the packing adapter if required

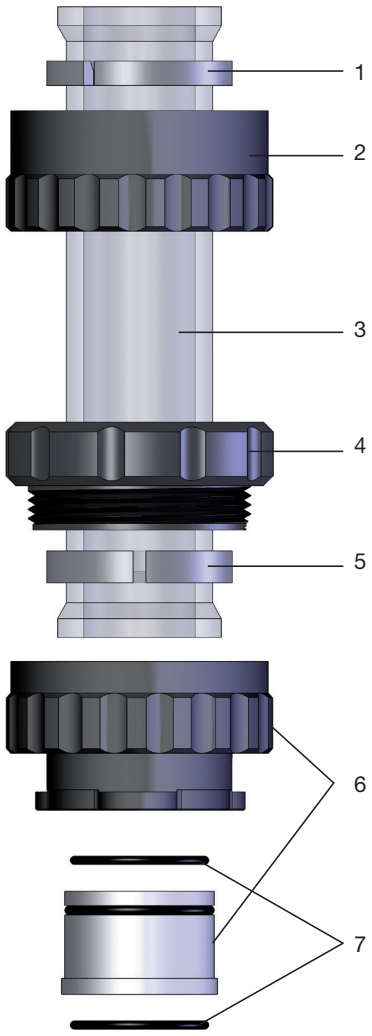
ATTENTION

Do not exceed the specified pressure limit when using the packing adapter to prevent it being from damaged. Please note the pressure limit of 5 bar when using a packing adapter.

Do not forget to insert the O-rings! The packing adapter should be firmly locked to prevent material getting trapped between the seals and glass body!

▶ Assembly of the packing adapter:

1. Assemble the packing tube and coupling unit. The counter nut (4) of the coupling unit is attached with the open end towards the end of the glass tube using the retaining ring (5).
2. At the other end of the packing tube the counter nut (2) and retaining ring (1) are attached to the glass body (3).
3. The nut of the coupling unit can now be fixed to the pre-assembled packing adapter. The glass body (3) is inserted.
4. The packing adapter is then attached to the column tube and locked in place by the counter nut on the column tube.
5. At the upper end of the packing adapter, the upper plunger from the glass column is inserted.
6. Now the packing adapter can be used to pack the column.



1 snap ring (column version)

2 counter nut (bayonet)

3 glass body (not included in column coupler)

4 column coupler counter nut

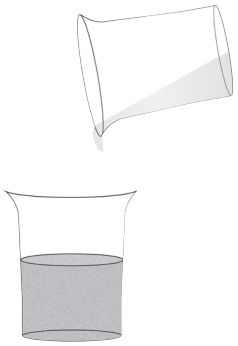
5 column coupler snap ring

6 connection screw (bayonet)

7 O-ring

Fig. 4.1: The packing adapter

4.2.2. Preparing the slurry



▶ Preparing the slurry:

1. Add the appropriate solvent or the buffer to the required amount of packing material (see information for the packing material)! The amount of packing material used determines the final height of the packed bed.

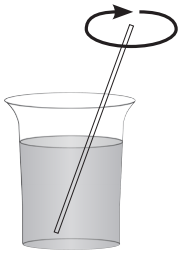
   Note: the total volume of the slurry must not exceed the maximum volume of the column plus packing adapter.

The packing is best performed using a slurry as dilute as possible. Always try to adjust the total volume of the slurry to the maximum volume of the column (plus packing adapter if used).

ATTENTION

Depending on the nature of the packing material a magnetic stirrer can cause mechanical damage to the material. Please ask the manufacturer of the packing material if you need more information.

2. Mix the slurry homogeneously with a spoon or spatula (or similar) avoiding air bubble formation.
3. Degas the homogeneous slurry in an ultrasonic bath.
4. Allow the slurry to settle. If particles (so-called fines) appear on the liquid surface, please remove them.



The removal of fines improves the properties of the packed column with respect to lower back pressures.

5. Before pouring the slurry into the column you should mix again gently to create a homogeneous suspension!

4.2.3. Packing the column

▶ **A. Slurry packing with applied flow**

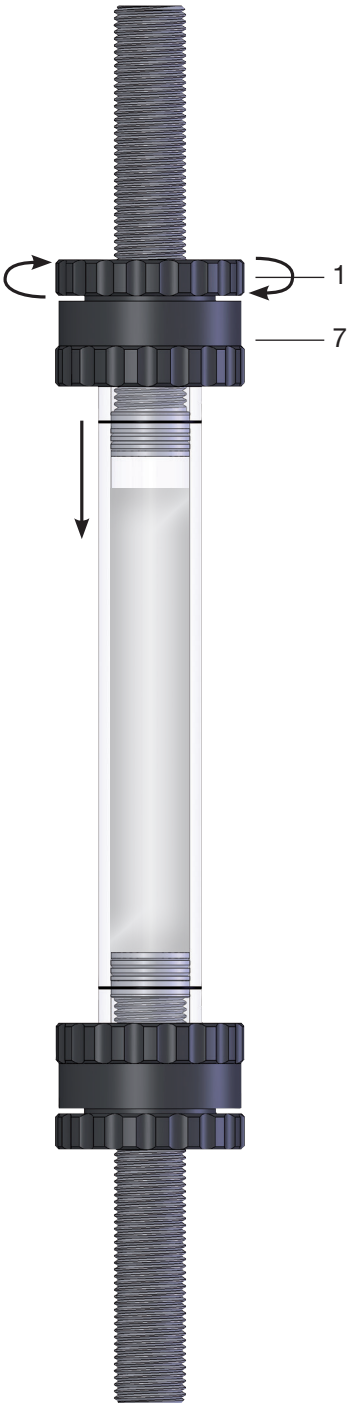
You can use this method to pack ECO^{PLUS} columns with rigid packing materials (e.g. silica- or alumina-based materials).

1. Make sure the column is exactly vertically to avoid the formation of dead volumes!
2. Pour the homogeneous, degassed slurry into the column quickly and without air bubbles. Let the slurry flow down the opposing glass wall. The column wall should then be rinsed with a minimal amount of solvent or buffer.

ATTENTION

Make sure that no particles get between the seal and glass wall.

3. Gently, but carefully insert the upper plunger and move it down into the column by turning the lock screw (1) counterclockwise.
4. Lower the plunger into the column until the first solvent exits through the upper plunger. Make sure that no air is left in the system.
5. As quickly as possible, connect a pump, remove the plug from the column outlet and then start the flow.
6. Slowly increase the flow rate to the desired pressure. At each increase in flow rate allow the solvent or the buffer to pass through the column until a constant pressure is reached. Only then should the flow rate (the pressure) be increased. The column bed will be visibly compressed.



The pressure is controlled up to the pressure limit of the column by the pump. If necessary the column should be packed with the maximum flow rate of the pump. The pressure limit depends both on the specification of the ECO^{PLUS} glass column, as well as the packing material used.

The flow rate used for packing should ideally be at least 20% higher than the flow rate used for subsequent separations.

7. After switching off the pump, the plunger should be moved by turning the counter screw (1) down until it touches the column bed. This should be done quickly and as evenly as possible without shaking.
8. Turn the pump on again and check for any dead volume. In accordance with paragraph 6, the upper plunger can be readjusted to the top of the column bed again after switching off the pump. This procedure should be repeated as many times as required until no more dead volume is present.
9. Finally, equilibrate the column with the desired eluent and test (see Chapter 4.2.4).

▶ B. Sedimentation process

ECO^{PLUS} columns can be packed with rigid gels (e.g., silica- or alumina-based particles) or soft gels (e.g. polymer gels or soft gels and agarose- or styrene-divinylbenzene-based particles) with this method.

1. With the column outlet sealed with a plug, pour the homogeneous and degassed slurry rapidly and as far as possible without air bubbles into the glass column. Add a small amount of solvent or buffer to the column to rinse the walls.
2. Allow the gel to settle (sedimentation). This can take several hours depending on the resin.

ATTENTION

The column packing must not run dry.

3. Drain the liquid to approx. 3 cm above the gel bed, by removing the plug at the outlet of the lower column.
4. Close the column outlet again with the plug to prevent the packing to running dry.

ATTENTION

Make sure that there are no particles between the seal and glass wall.

5. Gently, but carefully insert the upper plunger and move it down into the column by turning the lock screw.
6. Lower the plunger into the column until the first solvent exits through the upper plunger. Make sure that no air is left in the system.
7. The plunger is adjusted by turning the counter screw down until it touches the column bed. For pressure-labile materials no pressure must be generated by the plunger on the column bed. Please refer to the packing instructions from the manufacturer of the packing material.
8. Finally, equilibrate the column with the desired eluent and test it. ▶*Chapter 4.2.4., S. 31*.

4.2.4. Column test and acceptance criteria

The column test and the acceptance criteria should be designed with regard to the subsequent application. The preparation of the necessary specifications for the subsequent application of a method or the purification of a target compound requires customisation. The expected values for the achievable theoretical plate number can be found within the manufacturer specifications of the bulk media (see the example on this page).

Example for YMC-Triart Prep bulk media
After finishing the packing procedure, the theoretical plate number (N) and the peak symmetry should be evaluated.

Example conditions of column performance evaluation with YMC-Triart Prep C18-S:

Column size:	250 x 50 mm ID
Eluent:	methanol / water (85/15, v/v)
Flow rate:	50 mL/min
Detection:	UV at 254 nm
Sample:	1. Uracil (0.6 mg/mL) 2. Methyl benzoate (0.6 mg/mL) 3. Toluene (40 µL/mL)
Injection:	1 mL
Evaluation:	Theoretical plate number (N) of toluene

Expected theoretical plate number (N/m)*

	10 µm	15 µm	20 µm
C18	25,000	16,000	12,000
C8	23,000	15,000	11,000

** Values might be influenced by column or LC system*

For a successful packing 90% of the theoretical plate number should be reached (e.g. C18, 10 µm, 22,500) and the peak symmetry should be in between 0.7 and 1.3.

YMC Europe GmbH would be pleased to help you technically.

Contact:

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E-Mail: info@ymc.de

4.3. Packing adapter

ATTENTION

Do not exceed the specified pressure limit when using a packing adapter to prevent leakage. Please note the pressure limit of 5 bar when using a packing adapter.

The packing adapter is designed for non-pressure (sedimentation) packing of the column. However, low flow rates are possible, provided the pressure limit for the adapter is not exceeded. The use of a filler pipe when packing by means of sedimentation generally leads to the best results. Due to the increased maximum filling volume, the slurry concentration can be reduced and the packing process made easier. When packing with applied flow, the specification must be considered with respect to the pressure limits (see table on page 14).

Do not forget the O-rings! The packing adapter should be firmly locked to prevent material getting trapped between the seals and glass body!

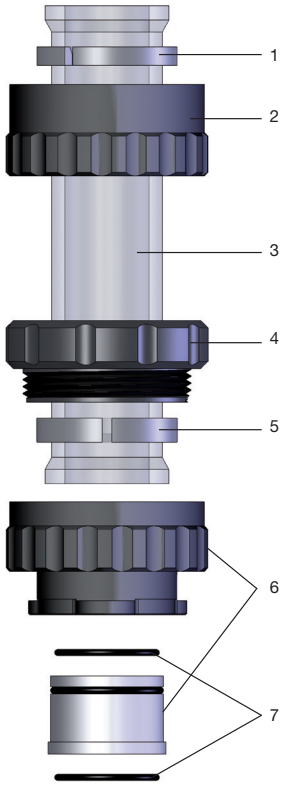


Fig. 5.1: Packing adapter

1 snap ring (column version)
2 counter nut (bayonet)
3 glass body (not included in column coupler)
4 column coupler counter nut
5 column coupler snap ring
6 connection screw (bayonet)
7 O-ring

▶ To assemble the packing adapter, please follow the numbered steps:

1. Assemble the packing tube and coupling unit. The counter nut (4) of the coupling unit is attached with the open end towards the end of the glass tube using the retaining ring (5).
2. At the other end of the packing tube the counter nut (2) and retaining ring (1) are attached to the glass body (3).
3. The nut of the coupling unit can now be fixed to the pre-assembled packing adapter. The glass body (3) is inserted.
4. The packing adapter is then attached to the column tube and locked in place by the counter nut on the column tube.
5. At the upper end of the packing adapter, the upper plunger from the glass column is inserted.
6. The packing adapter can now be used to pack the column.

For non-pressure packing (sedimentation) it is recommended to order and use a packing adapter from the list of parts in Table 6. The glass body of the packing adapter is not supplied as part of the packing adapter. This must be ordered with the required length separately.

Table 6: Packing adapter

Number	Article	Special features
1	Glass body (same ID as the column)	Must be ordered separately from the column coupler unit
1	ECO^{PLUS} coupling unit with Teflon insert (assembled)	
1	Column coupler counter screw with retaining ring	
1	Counter nut (bayonet) with retaining ring	
4	Viton® O-Ring	for AB Version
4	Kalrez® O-Ring	for SR Version

The part numbers of the ECO^{PLUS} column couplers are given in Table 7 and the ECO^{PLUS} glass bodies are listed in Table 8.

Table 7: ECO^{PLUS} coupling unit

Column ID [mm]	AB Version Coupling unit Part number	SR Version Coupling unit Part number
5	TAC05KU-AB	TAC05KU-SR
10	TAC10KU-AB	TAC10KU-SR
15	TAC15KU-AB	TAC15KU-SR
25	TAC25KU-AB	TAC25KU-SR
35	TAC35KU-AB	TAC35KU-SR
50	TAC50KU-AB	TAC50KU-SR

Table 8: ECO^{PLUS} glass bodies

Column ID [mm]	Max. bed length 125 mm Part number	Max. bed length 250 mm Part number	Max. bed length 500 mm Part number
5	TAC05/125-2	TAC05/250-2	TAC05/500-2
10	TAC10/125-2	TAC10/250-2	TAC10/500-2
15	TAC15/125-2	TAC15/250-2	TAC15/500-2
25	TAC25/125-2	TAC25/250-2	TAC25/500-2
50	TAC50/125-2	TAC50/250-2	TAC50/500-2

4.4. Information for Cleaning In Place (CIP)

The cleaning in place (CIP) procedure allows the cleaning of a packed ECO^{PLUS} column, without having to unpack it.

The cleaning of a packed chromatography column involves three steps:

- Regeneration of the column packing
- Sterilization
- Depyrogenation

Regeneration removes inorganic and organic contamination, which has become non-specifically bound to the chromatography matrix and substantially reduces the capacity and resolution of the column. This contamination is usually caused by lipids and pyrogens, protein aggregates, pigments, polyphenols or metal complexes.

Sterilization is the removal and/or denaturing of micro-organisms and spores that can contaminate the purified product. Most common method of treatment is the use of chemicals such as sodium hydroxide, acetic acid or ethanol.

Depyrogenation involves break-down of endotoxins that have become attached to the chromatographic packing material or to the column hardware (frits, tubing, etc.) and which can contaminate the product during purification. Often, the methods to sterilize the equipment destroy the pyrogens at the same time.

ATTENTION

Before cleaning the packed column with the CIP methods below, please check whether the column packing is compatible with the treatment. Otherwise, you will damage the stationary phase. Please also consider the version of the ECO^{PLUS} column being used (AB or SR).

Table 9: cleaning methods for CIP

Treatment	Contamination	Sterilisation	Depyrogenation
Pepsin, pH 1.5 - 2	Hydrolysis of adsorbed proteins	no	no
Non-ionic detergents (Triton X-100; Tween 80)	Removal of hydrophobic proteins and lipids	no	no
Cationic detergents, pH 9 - 11	Removal of hydrophobic proteins and lipids	no	partially
Urea, 6 - 8 M	Removal of protein aggregates	no	unknown
1 - 100 mM EDTA, pH depends on the stationary phase	Removal of metal complexes	no	no
0.1 - 1 M NaOH	In particular the removal of bonded hydrophobic proteins and lipopolysaccharides	good	good
0.5 - 1 M acetic acid in 60% ethanol	Removal of lipids, pigments, lipopolysaccharides and other lipophilic substances	very good	good
Organic solvents up to 100% (e.g. THF, isopropanol)	Removal of hydrophobic impurities	unknown	unknown

References:

De Bernardez Clark, E. (1998) Refolding of recombinant proteins. Current Opinion Biotechnol. 9, 157 - 163

Grönberg A et al. (2011) A tool for increasing the lifetime of chromatography resins. mABS, Vol 3, Issue 2, 192 - 202

Hagel L, Jagschies G, Sofer G, Cleaning and Sanitization In: Handbook of Process Chromatography–Development, Manufacturing, Validation and Economics. Second edition, London, UK: Academic Press 2008

Lilie, H., Schwarz, E. & Rudolph, R. (1998) Advances in refolding of proteins produced in E. coli. Current Opinion Biotechnol. 9, 497 - 501

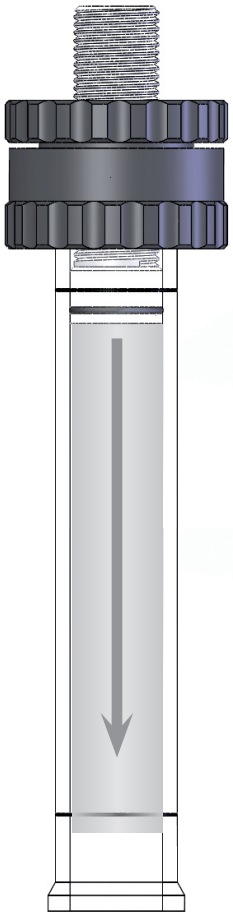
Romney AJD (1990) CIP: Cleaning In Place, 2nd edition, Int J Dairy Technol, Vol. 43, Issue 2, p. 59

Sofer GK, Nyström LE, Process Chromatography - A Guide to Validation, London, UK: Academic Press 1991

Tamime AY, Cleaning in Place: Dairy, Food and Beverage Operations. 3rd Edition, Oxford, UK: Backwell Publishing 2008

We recommend that you document all CIP processes. For this purpose, we have provided a log in the Appendix.
This can also be downloaded from http://ymc.de/ecoplus-glass-columns.html.

4.5. Unpacking



ATTENTION

Please do not dig out the packed material from the ECO^{PLUS} column with tools because the inner wall and the packing material will be damaged! We recommend the use of a pump to push the stationary phase.

Please collect the unpacked material in a container and dispose of it according to the manufacturer's instructions!

The solvents used should be disposed of in accordance with national standards.

▶ When unpacking the column please follow these steps:

1. Connect the column to the pump.
2. Remove the plunger from the outlet of the column and place a suitable container below the outlet.
3. Start the pump and push the material out from the open glass tube.

It is strongly recommend that the column is complete dismantled and the glass body cleaned after unpacking. (▶Chapter 3.4, Dismantling and Chapter 3.5, Cleaning).

5. Maintenance

If you need to replace any parts of the column, you will find the spare parts lists in ▶Chapter 9.1, spare parts list.

⚠ WARNING

Some chemicals used for cleaning can cause serious injury. The cleaning process for the chromatography columns should be performed in a fume cupboard or in a well-defined place.

ATTENTION

Before cleaning it is essential to check that the parts of the column are resistant to the chemicals used. Otherwise there is a risk of damage to the wetted parts. *(▶Chapter 3.8, Chemical resistance)*

ATTENTION

You should periodically clean the column otherwise you jeopardize the performance of the column. Check and replace the following parts if necessary (frits, glass body, O-ring seals).

5.1. Maintenance schedule

The specified maintenance plan here should be regarded as a general recommendation. The actual maintenance items and intervals should be designed depending on the frequency and type of use the column receives.

Interval	Maintenance
daily	• Check column bed height and tighten plunger sealing mechanism *(▶Chapter 3.6.1, Installing the plunger)*. • Check all tubes and connections for leaks and retighten fixing screws if necessary. Alternatively replace the corresponding tubes or connectors *(▶Chapter 3.6.2, Column to tubing connections)*.
every 3 months	• Check plunger rod and seals (O-rings or lamellae seals). If necessary replace the O-rings or plunger *(▶Chapter 3.4, Dismantling of the column)*.
every 6 months	• Check frit seating, if necessary replace the frit *(▶Chapter 5.2, Frit replacement)*

We recommend that you record all maintenance. For this purpose, we have provided a logbook in the Appendix.
This can also be downloaded from http://ymc.de/ecoplus-glass-columns.html.

5.2. Frit replacement

The frits can be changed for most column types using common laboratory equipment. Instructions are given in this manual for the various column types. However, frit replacement can also be carried out by YMC Europe GmbH if required, please contact us (see below).

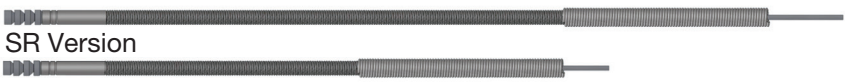
The part numbers for the corresponding frits can be found in ▶Chapter 9.1, Spare parts list.

5 mm ID columns

ATTENTION

The plungers for the 5 mm diameter columns in either the SR or AB version cannot be dismantled! The frit (PE/PTFE) is firmly attached to the stainless steel spindle. For this reason, the plunger can only be replaced as a complete unit. Spare component parts for this unit are not available.

AB Version



SR Version

10 – 50 mm ID SR version columns

Replacement of the frits in plungers from SR columns with 10-50 mm ID

The frits in plungers with an internal diameter of 10 - 50 mm have a trapezoidal body. This construction guarantees the safe retention of the frit in the plunger even at high loads. (▶Figure 5.1).

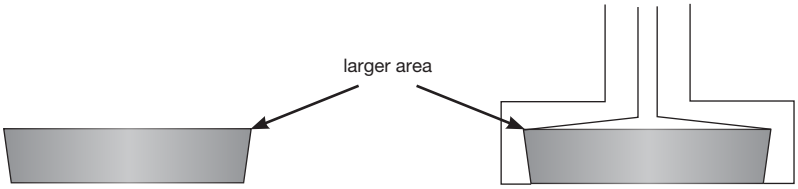


Fig. 5.1: Plunger with trapezoidal frit

▶ Change the frit please as the following steps:

1. Remove the old frit from the plunger, using the frit removal tool supplied. Ensure that the plunger is cleaned before replacing with a new frit.

⚠ CAUTION

Risk of burns!

2. Heat the SR plunger for about 3 hours at 121 °C in an oven (Material: PTFE).
3. Place a new cold frit on a smooth, clean surface with the side of the frit with the larger diameter facing up. Place the hot plunger with the opening over the frit and press down until the frit is fully located in the plunger.
4. Allow the plunger to cool prior to use, for between 1 hour (10 mm) and 3 hours (50 mm), depending on diameter.

10 – 15 mm ID AB version columns

Replacement of the frits in plungers from AB columns with 10 and 15 mm ID

The AB plungers are made of polyethylene, so the delicate thread can be damaged when overheated. Therefore, the end of the plunger must be heated in a water bath.

▶ To insert the frit please follow these instructions:

1. Remove the old frit from the plunger, using the frit removal tool supplied. Ensure that the plunger is cleaned before replacing with a new frit.

⚠ CAUTION

Risk of burns! Adjust the temperature of a suitable water bath to 60 – 70 °C.

2. Put the frit end of the plunger into an Erlenmeyer flask containing hot water. Add sufficient water so that the water level reaches up to the start of the thread.
3. Place the Erlenmeyer flask together with plunger into the prepared water bath.
4. After 10 minutes, the plunger is removed and a new, cold frit is securely placed in the hot plunger. The side of the frit with the larger diameter is placed into to the plunger first.
5. Allow to cool for at least 30 minutes.

25 – 50 mm ID AB version columns

Replacement of the frits in plungers from AB columns with 25 and 50 mm ID

The frit for plungers for columns having an internal diameter of 25 - 50 mm have a trapezoidal body. This construction guarantees the safe retention of the frit in the plunger even at high loads (▶Figure 5.1).

▶ To insert the frit please proceed in the following order:

1. Remove the old frit from the plunger, using the frit removal tool supplied. Ensure that the plunger is cleaned before replacing with a new frit.

⚠ CAUTION

Risk of burns!

2. Heat the AB plunger for about 1.5 hours at 105 °C in an oven (Material: PE).
3. Place a new cold frit on a smooth, clean surface with the side of the frit with the larger diameter facing up. Place the hot plunger with the opening over the frit and press down until the frit is fully located in the plunger.
4. Allow the plunger to cool prior to use, depending on the ID, for 2 hours (25 mm) or 3 hours (50 mm).

6. Troubleshooting guide

6.1. Troubleshooting table

Description of error	Possible reason	Remedial action
1. Dead volumes occur	Exceeding of the maximum allowable pressure for the column packing Pressure on the column packing	1. Switch off the pump 2. Turn column counter screw counterclockwise 3. Adjust the plunger onto the packing material surface. Do not force! ***ATTENTION*** **Danger of destroying the packing and the frit!**
2. Peak shape of the eluted substances deteriorates	1. Dead volume at the column inlet 2. Partially clogged entry frit 3. Partially clogged outlet frit 4. Separation performance of the stationary phase changed by contamination 5. Stationary phase mechanically destroyed	1. ▶Point 1 ”Dead volumes occur” 2. ▶Chapter 5.2 Frit replacement 3. ▶Chapter 5.2 Frit replacement 4. Rinse and consult the manufacturer of the packing material ▶Chapter 4.4, CIP 5. Replace packing material
3. Air (gas) in the column	Solvent has degassed or evaporated during storage	Recondition the column; maybe replacement packing is necessary
4. Abnormal pressure increase during use	1. Valve incorrectly switched 2. Blocked frit 3. Fittings over tightened	1. Check valve positions 2. ▶Chapter 5.2 Frit replacement 3. Renew fixing screws and ferrules, re-cut the end of the tube
5. Pressure drop during use	1. Leak in the tube pipe or fittings between the pump and column 2. Solvent reservoir empty	1. Check tubing and connections 2. Refill solvent
6. Solvent drips from the column	1. Plunger leaking 2. Connection tube leaking	1. Visual inspection, is solvent passing the seal? Tighten the screw nut again, remove plunger -> clean glass body and seals -> insert new plunger/ replace O-rings -> recondition column 2. Tighten fixing screw -> check connecting tubing for leaks –> recondition column

7. Storage, packaging and transport

7.1. Storage

ATTENTION

Store the glass column body securely! This will avoid the column falling and the glass breaking.

For storage of packed columns, we recommend that you consider the chemical resistance of the wetted materials (▶Chapter 3.8, Chemical resistance), and chemical resistance of the packed stationary phase (please check with the manufacturer).

7.2. Packaging and transport

ATTENTION

Damage can occur with improper packaging at a high altitude.

We encourage you to always use the original packaging for transportation (▶Chapter 2.2, Delivery inspection). If you no longer have the original packaging, please pack the column in a similar manner. For this purpose, please make sure the chromatography column or other component parts are not in direct contact with the outer packaging. Allow at least 50 mm distance between the chromatographic column and packaging by using foam padding (eg polystyrene, etc.) as a spacer.

- Attach appropriate warnings (e.g. “Caution Glass”).

8. Disposal

To dispose of the ECO^{PLUS} column, it must be dismantled and each part disposed of according national and other applicable rules for environmentally safe disposal.

9. Spare and replacement parts

9.1. Spare parts list

ECO^{PLUS} glass bodies

Part No.	ID [mm]	Max. bed length [mm]
TAC05/125-2	5	125
TAC05/250-2	5	250
TAC05/500-2	5	500
TAC10/125-2	10	125
TAC10/250-2	10	250
TAC10/500-2	10	500
TAC15/125-2	15	125
TAC15/250-2	15	250
TAC15/500-2	15	500
TAC25/125-2	25	125
TAC25/250-2	25	250
TAC25/500-2	25	500
TAC35/125-2	35	125
TAC35/250-2	35	250
TAC35/500-2	35	500
TAC50/125-2	50	125
TAC50/250-2	50	250
TAC50/500-2	50	500

ECO^{PLUS} O-ring, EPDM, for AB columns, 2 pieces per pack

Part No.	ID [mm]
TAC05/04E/P2	5
TAC10/04E/P2	10
TAC15/04E/P2	15
TAC25/04E/P2	25
TAC35/04E/P2	35
TAC50/04E/P2	50

ECO^{PLUS} O-ring, Kalrez® white, for SR columns, 2 pieces per pack

Part No.	ID [mm]
TAC50/04K-NK/P2	50

Frits, 2 pieces per pack

Part No.	ID [mm]	Porosity [µm]
Polyethylene frits		
TAC05PE10/P2	5	10
TAC05PE5/P2	5	5
TAC10PE10-2/P2	10	10
TAC10PE5-2/P2	10	5
TAC15PE10-2/P2	15	10
TAC15PE5-2/P2	15	5
TAC25PE10-2/P2	25	10
TAC25PE5-2/P2	25	5
TAC35PE10-2/P2	35	10
TAC35PE5-2/P2	35	5
TAC50PE10-2/P2	50	10
TAC50PE5-2/P2	50	5

Part No.	ID [mm]	Porosity [µm]
Sintered glass frits		
TAC05G10/P2	5	10
TAC05G2/P2	5	2
TAC10G10-2/P2	10	10
TAC10G2-2/P2	10	2
TAC15G10-2/P2	15	10
TAC15G2-2/P2	15	2

Part No.	ID [mm]	Porosity [µm]
Stainless steel frits		
TAC25S10-2/P2	25	10
TAC25S2-2/P2	25	2
TAC35S10-2/P2	35	10
TAC35S2-2/P2	35	2
TAC50S10-2/P2	50	10
TAC50S2-2/P2	50	2

Plunger, Polyethylene, for AB columns (incl. O-Ring, without frit)

Part No.	ID [mm]
Short plunger	
TAC05/03STS-AB-4	5
TAC10/03STS-AB-2	10
TAC15/03STS-AB-2	15
TAC25/03STS-AB-2	25
TAC35/03STS-AB-2	35
TAC50/03STS-AB-2	50

Part No.	ID [mm]
Long plunger	
TAC05/03STL-AB-4	5
TAC10/03STL-AB-2	10
TAC15/03STL-AB-2	15
TAC25/03STL-AB-2	25
TAC35/03STL-AB-2	35
TAC50/03STL-AB-2	50

Plunger, PTFE, for SR columns (without frit)

Part No.	ID [mm]
Short plunger	
TAC05/03STS-4	5
TAC10/03STS-3	10
TAC15/03STS-3	15
TAC25/03STS-3	25
TAC35/03STS-3	35
TAC50/03STS-NK	50

Part No.	ID [mm]
Long plunger	
TAC05/03STL-4	5
TAC10/03STL-3	10
TAC15/03STL-3	15
TAC25/03STL-3	25
TAC35/03STL-3	35
TAC50/03STL-NK	50

Accessories for glass columns

Part No.	Description
FA	frit removal tool
K.P316-5	plugs PTFE (1/4 inch-28G), 5 pieces per pack
K.P620-5	coupling unit (double-ended ID 1/4 inch-28G), 5 pieces per pack
K.P621-5	adapter 1/4 inch-28G to M6, 5 pieces per pack
K.P627	adapter 10-32 to 1/4 inch-28G (incl. 1 F-300 fitting for 1/16 inch tubing)

Tubing

Part No.	Description
PT0,8FE1,6/M5	FEP tubing 0.8 x 1.6 mm (1/16 inch), 5 m per pack
PT1,6FE3,2/M5	FEP tubing 1.6 x 3.2 mm (1/8 inch), 5 m per pack
JR-T-082-M3	Tefzel tubing 1/16 inch (for ID 5 mm), 3 m per pack

Fittings (fixing screw and ferrules)

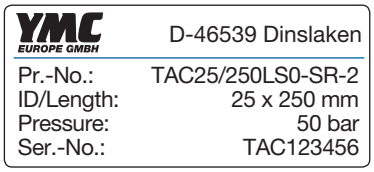
Part No.	Description
JR-55050-10	Fixing screw, 1/4 inch-28, PPS, for collapsible ferrule for 1/16 inch tubing, 10 pieces per pack
JR-CL-CB1KF	Ferrule, collapsible, for 1/16 inch tubing, 10 pieces per pack
K.P201X	Fixing screw 1/4 inch-28G for 1/16 inch tubing, 10 pieces per pack
K.P200NX	Ferrule for 1/16 inch tubing, 10 pieces per pack
K.P207X	M6 fixing screw, for 1/16 inch tubing, 10 pieces per pack
K.P200NX	M6 Ferrule, for 1/16 inch tubing, 10 pieces per pack
JR-55051-10	Fixing screw, 1/4 inch-28, PPS, for collapsible ferrule for 1/8 inch tubing, 10 pieces per pack
JR-CL-CB2KF-10	Ferrule, collapsible, for 1/8 inch tubing, 10 pieces per pack
K.P301X	Fixing screw 1/4 inch-28G for 1/8 inch tubing, 10 pieces per pack
K.P300NX	Ferrule for 1/8 inch tubing, 10 pieces per pack
K.P307X	M6 fixing screw, for 1/8 inch tubing, 10 pieces per pack
K.P300NX	M6 Ferrule, for 1/8 inch tubing, 10 pieces per pack

Part No.	Description
JR-58000-5	10-32 fixing screw/ferrule for 1/16 inch tubing, 5 pieces per pack

Part No.	Description
JR-797	Clean-Cut tubing cutter

9.2. Column versions available

The ECO^{PLUS} columns are available in different versions. These versions are derived from the combinations of the inner diameter, the maximum bed length, the stamp type of frit material, the frit porosity and the column variant (AB or SR). Any combination gives an individual order number, from which the column parameters are derived. The order number can be found on the label on the column glass body. The label also contains information about the maximum pressure that can be exposed to the column.



In Table 9, the column parameters are listed, from which the different part numbers (column versions) are generated. Their combination (from left to right) gives the part number. For a better understanding in the lower half of the table is an example of the generation of the Part No. TAC25/500S2-SR-2. This contains the information for a ECO^{PLUS} column with a maximum bed length of 500 mm, 2 short plungers and stainless steel frits with a porosity of 2 microns. For this purpose, the column is solvent resistant (SR version).

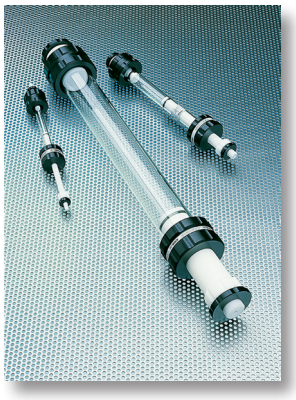
In the flyer (ECO^{PLUS} laboratory glass columns) which is available to download from ..... you can find detailed information on all the columns versions available.

Table 9: Column versions and part numbers

Combination possibilities	**TAC05/** (5 mm ID) **TAC10/** (10 mm ID) **TAC15/** (15 mm ID) **TAC25/** (25 mm ID) **TAC35/** (35 mm ID) **TAC50/** (50 mm ID)	**125** (125 mm max. bed length) **250** (250 mm max. bed length) **500** (500 mm max. bed length)	**—** (Standard Version has 2 short plungers - no letter is inserted) **SL** (1 short/1 long plunger) **L** (2 long plungers)	**PE** Polyethylene (AB Version) **G** Sintered glass (SR Version with ≤ 15 mm ID) **S** Stainless steel (SS) (SR Version with ≥ 25 mm ID)	**2** (2 µm) **5** (5 µm, only Polyethylene Frits) **0** (10 µm)	**-AB-2** (aqueous buffer) **-SR-2** (solvent resistant)
Example Co. No. (part number)	**TAC25/**	**500**		**S**	**2**	**-SR-2**
ID	25 mm					
Max. bed length		500 mm				
Plunger type			Standard version			
Frit material				Stainless steel		
Frit porosity					2 µm	
Version						SR version

On request, special versions of the columns can be manufactured. YMC Europe GmbH would be happy to discuss your requirements.

10. Product cross-references

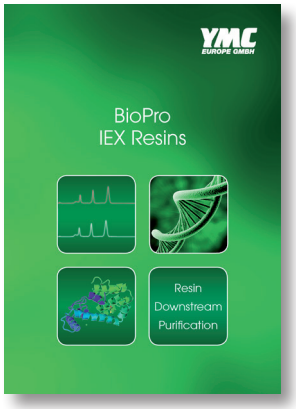


ECO glass columns:

- Cost effective alternative to ECO^{PLUS} glass columns
- Range of bed lengths 0 – 1000 mm
- Variable bed volume up to 5 Liter
- Robust construction
- Pressure range up to 30 bar
- FDA, BSE/TSE certificates available

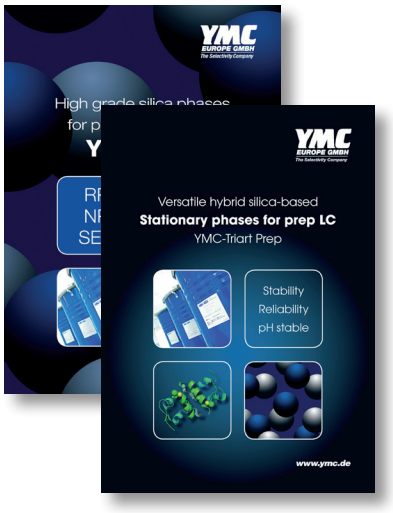
Pilot columns:

- for the scale up to process scale
- 60 mm to 450 mm inner diameter
- biocompatible AB-version
- SR-version with plungers of stainless steel available
- custom sizes on demand
- FDA certification, declaration of freedom from TSE/BSE, USP Class VI certification



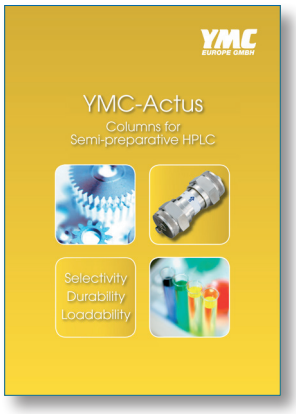
BioPro:

- polymeric ion exchange material for biochromatography
- available as strong anion- or cation-exchanger
- excellent flow properties and high dynamic binding capacity
- extremely economic for industrial scale applications
- particle sizes: 10 µm; 20 µm; 30 µm; 75 µm



YMC*Gel / YMC-Triart Prep:

- general purpose preparative phases on high grade silica base
- available as silica or with C18, C8, C4, C1, phenyl, cyano, amino or diol bonding
- particle sizes: 10 µm; 15 µm; 20 µm; 50 µm
- pore sizes: 8 nm; 12 nm; 20 nm; 30 nm
- YMC-Triart Prep: pH- and temperature- stable hybrid material
- robust, flexible and economic



YMC-Actus:

- columns packed for high pressure applications
- dynamic axial compression for an improved chromatographic performance
- robust performance for easy handling with complete reliability
- inner diameters from 20 mm to 1000 mm
- lengths from 250 mm to 1000 mm

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11. Index

12. Appendix

Logbook

Activity No.	Date	Activity (e.g. maintenance, cleaning, etc.) and / or error description	Action taken	Con-ducted by	Result / commentary

Activity No.	Date	Activity (e.g. maintenance, cleaning, etc.) and / or error description	Action taken	Con-ducted by	Result / commentary

Activity No.	Date	Activity (e.g. maintenance, cleaning, etc.) and / or error description	Action taken	Con-ducted by	Result / commentary

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