ÄKTAxpress

Take advantage of your routines; be in two places at once

Keeping pace with today's purification challenges

ÄKTAxpress™ is a complete, intelligent, and robust solution for routine, unattended purification of affinity-tagged proteins and antibodies.

Automated multistep purification is the optimal way to efficiently achieve the highest possible protein purity. In addition, ÄKTAxpress can run multiple samples, thus providing both the required purity and throughput.

It is possible to purify proteins expressed at all levels, as long as there is at least 2 mg of the target protein in the sample. Dual-tagged proteins can be purified easily and you have the option of on-column tag cleavage for all protocols that starts with an affinity or ion exchange step.

A complete solution

ÄKTAxpress has a modular design that allows for a high degree of adaptability. A Single or Twin system can be upgraded with separate modules, and up to 12 modules can be controlled from one computer. Each module is run independently, and different protocols can be run even though a single computer controls them. Each module can purify up to four different proteins serially in one run.

The software enables the detection and storage of intermediate peaks in capillary loops, allowing the system to automatically wash, equilibrate, elute, or re-inject samples on the different columns connected to the system.

An extensive range of prepacked columns for purification of tagged proteins and antibodies are designed to work together with the system. UNICORN™ software allows easy system set-up and unattended protein purification, with full documentation and evaluation.

The method wizard makes it easy to create a method plan for a purification protocol. It guides you with checkboxes and pull down menus, making it simple to create protocols with up to four different purification steps. Simply decide the number of purification steps and choose the appropriate columns from the column list. There are pre-programmed default values for all running parameters, which minimizes the need for chromatography expertise. An advanced zone in the method wizard gives you the option to change the default settings.

Robust algorithms and combinations of watch commands make it possible for the system to automatically process interesting protein peaks at different stages in the purification process.

ÄKTAxpress Single

Start with one ÄKTAxpress module and grow—
Up to 12 modules can be controlled from one computer. Each module has a fixed configuration that consists of the system pump, an air sensor, a UV detector (280 or 254 nm), a conductivity detector and an integrated fraction collector for 24- or 96-deepwell plates. All necessary items, such as valves, sample loops and tubing, are pre-installed. An optional flask holder is available.

ÄKTAxpress Twin

Intelligence
Hardware, software and media designed to work together; no preknowledge about the target protein's elution profile is needed

Unattended use
Ability to automatically run up to four samples per system through multistep purifications with minimal hands-on time

Output
Yields of 1 to over 50 mg of pure protein per sample are easily produced

Purity
Automated multistep purification provides target protein purity >95% without manual interactions

Flexibility
Different protocols can be run simultaneously on different modules

Reliability
Well established service and support experience with over 20 000 installed ÄKTAdesign systems
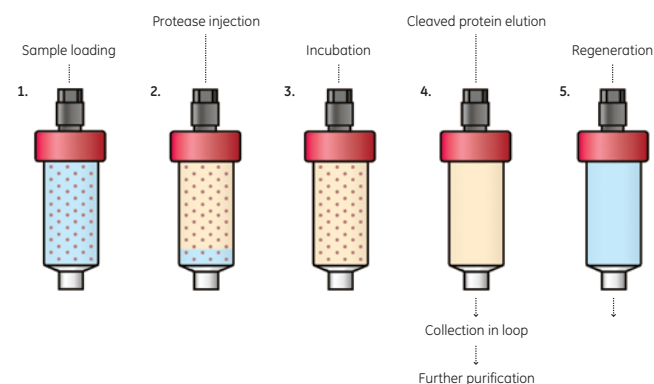
Tagged protein purification – high purity for structural and functional studies

ÄKTAxpress can handle protocols with up to four purification steps and optional tag removal. The methods used are: affinity (AC), desalting (DS), ion exchange (IEX), and gel filtration (GF).

Dual-tagged proteins can be purified using protocols that include two AC steps. Flexible peak selection enables successful purification of proteins expressed at low levels.

Automated tag removal

An optional on-column tag removal step can be integrated in all protocols that start with AC or IEX. The method works for a broad range of proteases, both untagged and tagged.

Sample loading · Protease injection · Incubation · Cleaved protein elution · Regeneration

1. 2. 3. 4. 5.

Collection in loop

Further purification

Supported protocols for tagged protein purification

Protocol	Typical run time (h)
One step – Max. four samples per module	
AC	2.5
DS	0.5
IEX	2.5
GF	14
Two steps – Max. four samples per module	
AC-DS	4.5
AC-GF	18
DS-AC	3
DS-IEX	3
IEX-DS	4.5
IEX-GF	18
Three steps – Max. three samples per module	
AC-(DS)-AC	7
AC-(DS)-IEX	7
IEX-(DS)-AC	7
IEX-(DS)-IEX	7
DS-AC-DS	4
DS-AC-GF	11
DS-IEX-DS	4
DS-IEX-GF	11
Four steps – Max. two samples per module	
AC-(DS)-AC-DS	5.5
AC-(DS)-AC-GF	12.5
AC-(DS)-IEX-DS	5.5
AC-(DS)-IEX-GF	12.5
IEX-(DS)-AC-DS	5.5
IEX-(DS)-AC-GF	12.5
IEX-(DS)-IEX-DS	5.5
IEX-(DS)-IEX-GF	12.5

(DS) = optional desalting step

Save time and effort

The system is designed to eliminate the manual steps involved in multistep purifications.

	Traditional methods	With ÄKTAxpress
When working with tagged proteins		
Sample preparation	Harvest your cells, sonicate, clarify sample, purify.	Using HisTrap FF crude – apply unclarified sample directly on column. Save up to 1 h/sample.
Running multistep protocols	Pool fractions between each step, start next purification step and apply sample to the column.	Automatic sample loading of collected peak onto next column. Save up to 12 h when running a four-step protocol.

Skip a step in sample prep

When using the HisTrap™ FF crude column, you can apply unclarified cell extract directly onto the column and save 40 to 80 min. per sample without risking any clogging of the column.

Without HisTrap FF crude
Cell culture > Sonication > Sample prep > Load sample > Purified protein
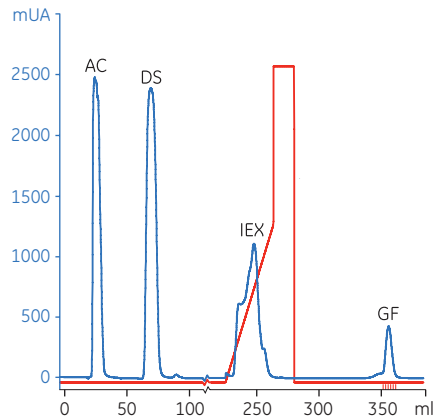
With HisTrap FF crude
Cell culture > Sonication > Load sample > Purified protein

ÄKTAxpress results for tagged protein purification

Four-step purification

(AC-DS-IEX-GF) of APB7, a histidine-tagged protein of M_r 28 000
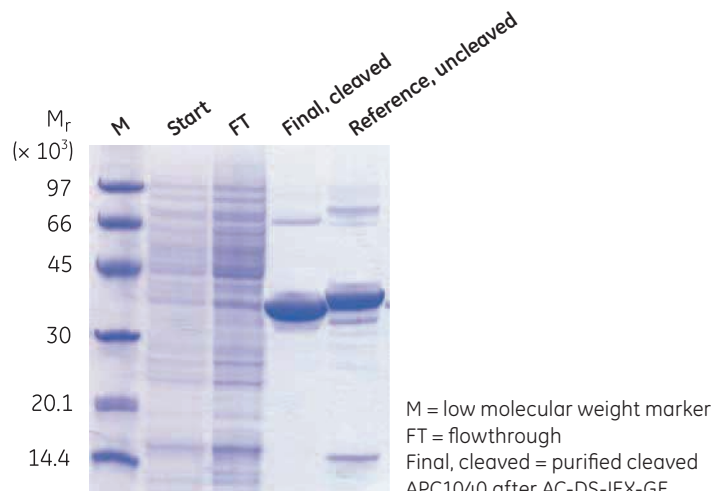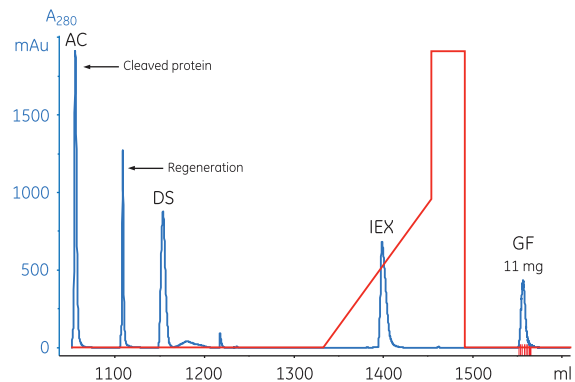
AC: HisTrap HP, 5 ml
DS: HiPrep 26/10 Desalting
IEX: 2 × HiTrap Q HP, 1 ml
GF: HiLoad 16/60 Superdex 75 pg

Four-step purification and online tag removal included of APC1040, a histidine-tagged protein ($M_r = 38\ 900$) with a TEV cleavage site

AC: HisTrap HP, 5 ml
DS: HiPrep 26/10 Desalting
IEX: RESOURCE Q, 6 ml
GF: HiLoad 16/60 Superdex 75 PG

Protease: AcTEV histidine-tagged

M = low molecular weight marker
FT = flowthrough
Final, cleaved = purified cleaved APC1040 after AC-DS-IEX-GF

## Purification of a double affinity-tagged protein

Dual-tagged proteins enable more targeted affinity purification of full length proteins. If the tags are at different terminals of the protein a full length protein is also secured. Protocols containing two affinity steps have been developed to support purification of dual-tagged proteins.

In this application, green fluorescent protein (GFP) tagged with histidine at its N-terminus and GST at its C-terminus was purified in three different ways:

A.   AC(His)-DS for purification by the histidine tag
B.   AC(GST)-DS for purification by the GST tag
C.   AC(His)-DS-AC(GST)-DS for purification by both tags, first by the histidine tag and then by the GST tag
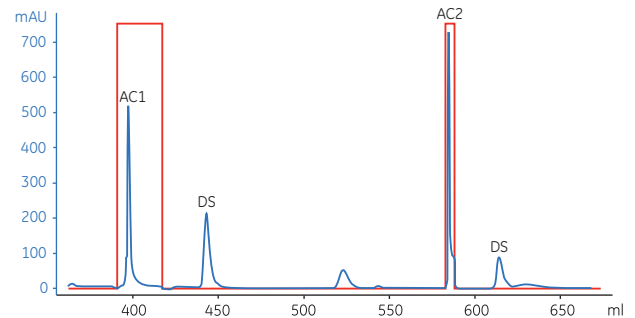
AC1:	HisTrap HP, 5ml
DS:	HiPrep 26/10 Desalting
AC2:	GSTrap 4B, 1ml
DS:	HiPrep 26/10 Desalting
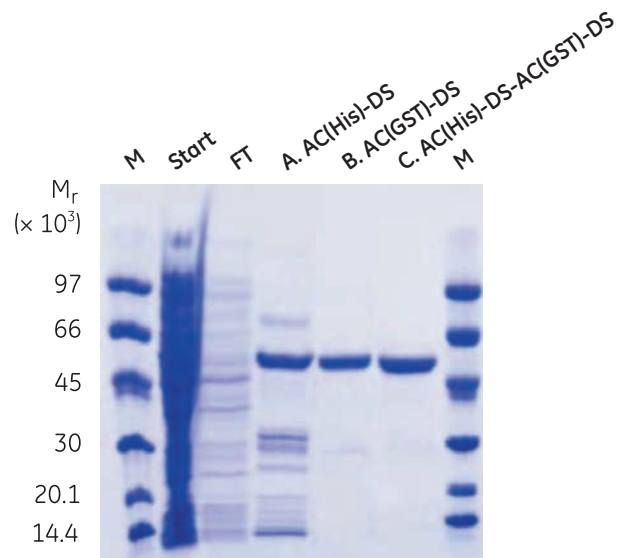
M = low molecular weight marker
FT = flowthrough after AC1

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Antibody purification – high purity for preclinical screening

Supported protocols for antibody purification

Protocol	No. of samples/module
AC	4
AC-DS using one single desalting column for all samples	4
AC-DS with individual desalting columns for each sample	2
AC-GF using one single gel filtration column for all samples	4
AC-GF with individual gel filtration columns for each sample	2

Note: A separate affinity column is used for each sample.

Alkali stable HiTrap MabSelect SuRe
The same cleaning procedure using 0.5 M NaOH is applicable for both the system and column without loss of binding capacity.
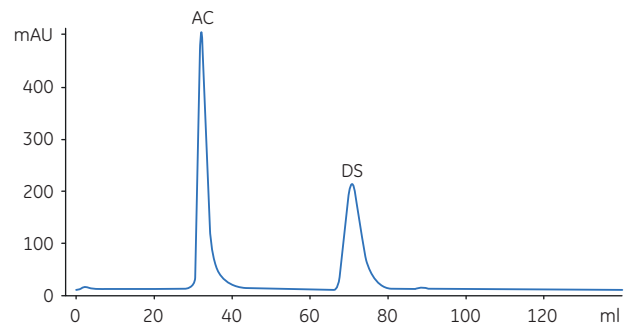
Automated neutralization
Choosing the desalting step automatically places the MAb in the appropriate buffer, avoiding degradation of the monoclonal antibody.

ÄKTAxpress results for monoclonal antibodies

Two-step AC-DS of human IgG$_1$ from hybridoma cell culture supernatant.

AC: HiTrap MabSelect SuRe 5 ml
DS: HiPrep 26/10 Desalting

Automated cleaning procedure between samples

Extended washing procedures enable automatic processing of a large number of samples and minimize the risk of cross-contamination.

One point of control

The ÄKTAxpress system is controlled by UNICORN version 5.11 or higher. Algorithms and combinations of watch commands have been developed and optimized to enable intelligent automation and robust peak handling functions. Now even the most difficult of proteins can be tackled with ÄKTAxpress.

The method wizard guides you through each stage of the method programming process. A method plan is generated, based on the answers to a series of questions. Columns are selected from the column library – which includes all the prepacked columns designed to work with the intended application. Optimized values for all running parameters are used as a default to make it easy to create new method plans. An advanced zone within the wizard makes it possible to change default settings to further optimize the purification of your samples.

1 Easy to create a method

When you enter the main menu of the wizard, you can choose to make a method plan for a purification protocol and/or different maintenance activities.
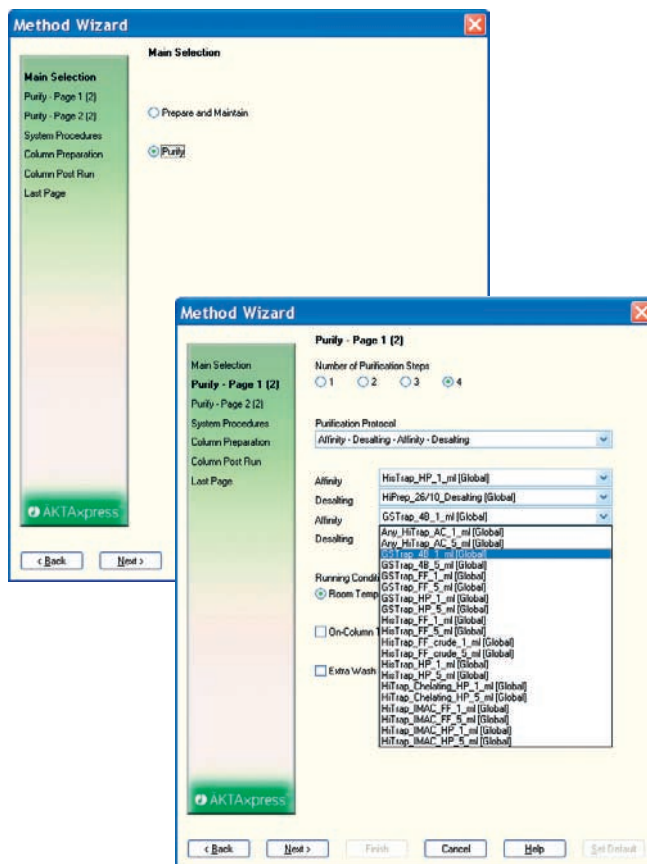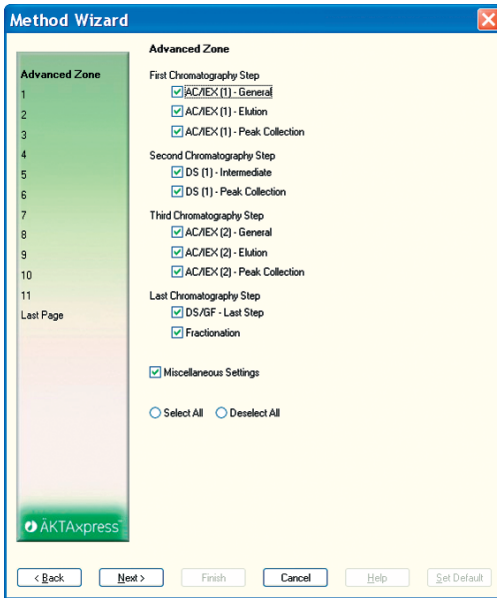

2 Pick what you want

To prepare a purification method plan, all you need to do is select what purification protocol you want to run and choose the appropriate columns from the column list. The rest is up to the wizard.

3 More advanced selection

You can adjust running parameters in the advanced zone of the wizard if needed.



4 Bar code data entry

When starting a run, you assign the method plan to your different system modules and enter the number of samples you want to run per module. Sample data can be entered either manually or by using a bar code reader.


5 Data in real time; monitor what's happening on all modules

You get data in real time while you run the method, and you can follow what is happening in detail on one module or have an overall status of all running modules.
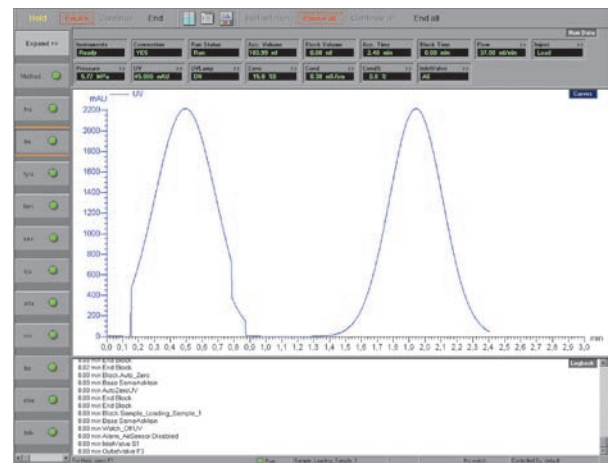
6 Evaluation and creating reports

Results are displayed in a way that makes evaluation simple. Reports are easily created by using the optimized formats.
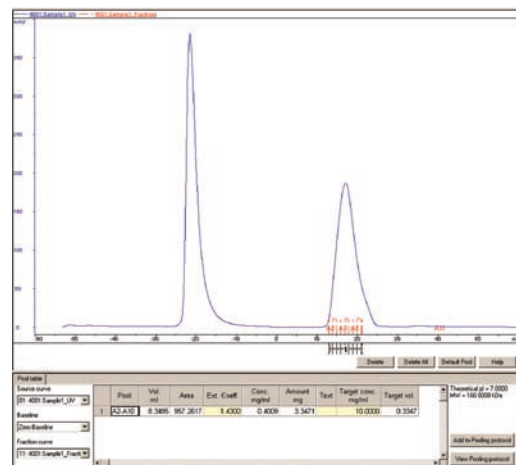
Ordering information

ÄKTAxpress

ÄKTAxpress Single system	28-9089-36
ÄKTAxpress Twin system	11-0012-84
ÄKTAxpress module for system extension	18-6645-01
ÄKTAxpress software (includes strategy for tagged proteins and antibodies)	28-9053-45

Related products

Gel filtration

HiLoad™ 16/60 Superdex™ 75 pg, 1 × 120 ml	17-1068-01
HiLoad 16/60 Superdex 200 pg, 1 × 120 ml	17-1069-01
HiLoad 26/60 Superdex 75 pg, 1 × 320 ml	17-1070-01
HiLoad 26/60 Superdex 200 pg, 1 × 320 ml	17-1071-01
HiPrep™ 16/60 Sephacryl S-100 HR, 1 × 120 ml	17-1165-01
HiPrep 16/60 Sephacryl S-200 HR, 1 × 120 ml	17-1166-01
HiPrep 16/60 Sephacryl S-300 HR, 1 × 120 ml	17-1167-01
HiPrep 26/60 Sephacryl S-100 HR, 1 × 320 ml	17-1194-01
HiPrep 26/60 Sephacryl S-200 HR, 1 × 320 ml	17-1195-01
HiPrep 26/60 Sephacryl S-300 HR, 1 × 320 ml	17-1196-01

Desalting

HiPrep 26/10 Desalting, 1 × 53 ml	17-5087-01
HiPrep 26/10 Desalting, 4 × 53 ml	17-5087-02
HiTrap™ Desalting, 5 × 5 ml	17-1408-01

Affinity chromatography

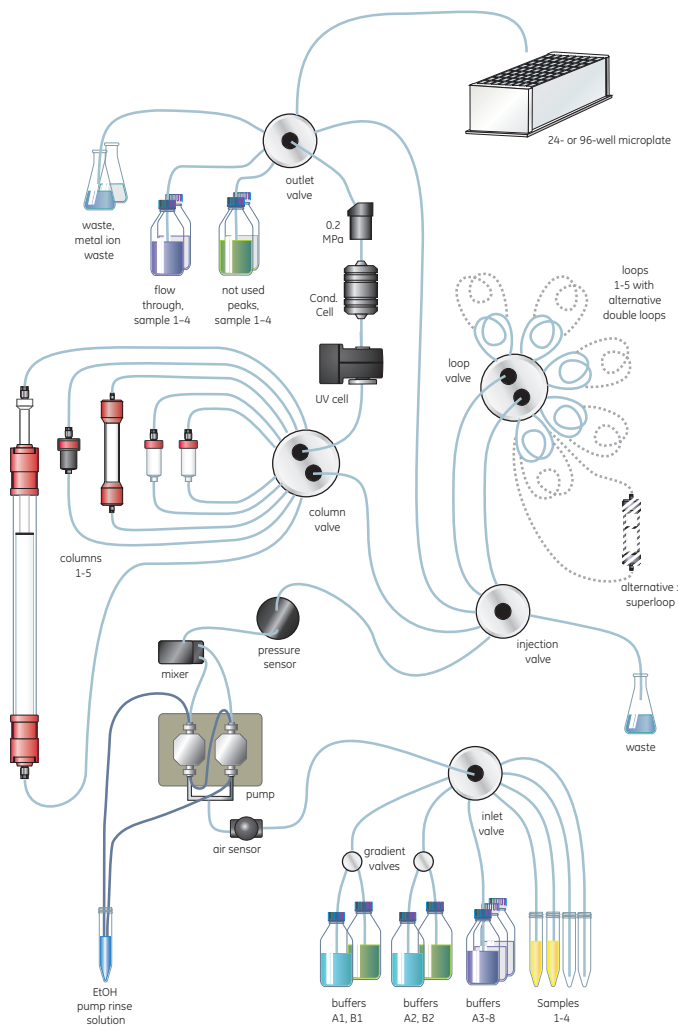|	
HisTrap HP, 5 × 1 ml	17-5247-01
HisTrap HP, 5 × 5 ml	17-5248-02
HiTrap MabSelect SuRe™ 5 × 1 ml	11-0034-93
HiTrap MabSelect SuRe 1 × 5ml	11-0034-94
HiTrap MabSelect SuRe 5 × 5 ml	11-0034-95
HiTrap Chelating HP, 5 × 1 ml	17-0408-01
HiTrap Chelating HP, 5 × 5 ml	17-0409-03

Related products (cont.)

GSTrap™ HP, 5 × 1 ml	17-5281-01
GSTrap HP, 5 × 5 ml	17-5282-02
GSTrap FF, 5 × 1 m	17-5130-01
GSTrap FF, 5 × 5 ml	17-5131-02
GSTrap 4B, 5 × 1 ml	28-4017-45
GSTrap 4B, 5 × 5 ml	28-4017-48
MBPTrap™ HP, 5 × 1 ml	28-9187-78
MBPTrap HP, 1 × 5 ml	28-9187-79
MBPTrap HP, 5 × 5 ml	28-9187-80
StrepTrap™ 5 × 1 ml	28-9075-46
StrepTrap 1 × 5 ml	28-9075-47
StrepTrap 5 × 5 ml	28-9075-48

Ion exchange chromatography

HiTrap SP HP, 5 × 1 ml	17-1151-01
HiTrap SP HP, 5 × 5 ml	17-1152-01
HiTrap Q HP, 5 × 1 ml	17-1153-01
HiTrap Q HP, 5 × 5 ml	17-1154-01
RESOURCE™ Q, 1 × 1 ml	17-1177-01
RESOURCE Q, 1 × 6 ml	17-1179-01
RESOURCE S, 1 × 1 ml	17-1178-01
RESOURCE S, 1 × 6 ml	17-1180-01
Mono Q™ 5/50 GL, 1 × 1 ml	17-5166-01
Mono S™ 5/50 GL, 1 × 1 ml	17-5168-01

Accessories

Large column holder	28-4007-37
Flask holder	18-1177-79
HiPrep 16/xx Column Wrap	28-9021-50
Loop extension kit for ÄKTAxpress	28-9044-38
Superloop™, 1/16" fittings (ÄKTAdesign™) 10 ml	18-1113-81
Superloop, 1/16" fittings (ÄKTAdesign) 50 ml	18-1113-82

24- or 96-well microplate

outlet valve

waste, metal ion waste

flow through, sample 1–4

not used peaks, sample 1–4

0.2 MPa

Cond. Cell

UV cell

loops 1-5 with alternative double loops

loop valve

columns 1-5

column valve

alternative : superloop

pressure sensor

injection valve

mixer

pump

waste

air sensor

EtOH pump rinse solution

gradient valves

inlet valve

buffers A1, B1

buffers A2, B2

buffers A3-8

Samples 1-4

ÄKTAxpress – System specifications

System flow rate	0.1–65 ml/min
Flow accuracy rate	± 2%
Operating pressure	3 MPa (30 bar, 435 psi)
UV wavelength range	280 or 254 nm
UV flow cell length	2 mm
Conductivity range	1 µS/cm to 999.9 mS/cm
Solvent compatibility	All commonly used chromatographic solvents
Module size	250 mm × 700 mm × 650 mm (W × H × D)
Weight	27 kg/module

A schematic flow diagram for one module set up to run a four step protocol with two samples. The Superloop is used to hold the protease solution used for tag cleavage or the start protein when being repetitively added (e.g., to a gel filtration column).

For contact information for your local office,
please visit: www.gelifesciences.com/contact

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imagination at work