# Ion exchange chromatography

Ion exchange chromatography (IEX) is a very useful technique that separates proteins on the basis of differences in their net surface charge in relation to the pH of the surroundings. Every protein has its own charge/pH relationship.

WorkBeads ion exchangers are also ideal for peptides, viruses, vaccines and oligonucleotides. They show excellent results for larger peptides, particularly insulin. Two different bead sizes are available for optimal purity in all steps of a purification process (capture, enhancement and polishing).



### Cation exchangers



### WorkBeads 40S WorkBeads 100S

- Designed for research and industrial scale purifications
  of proteins and peptides
- Two different bead sizes, 45 μm and 100 μm
- High chemical stability for easy cleaning-in-place
- High binding capacity also during high flow rates
- Available in several different GoBio prepacked columns



Structure of the ligand used in WorkBeads 40S

### Applications

#### Separation using GoBio Prep 16x100 40S

Column:	GoBio Prep 16x100 40S (20 mL)
Flow rate:	150 cm/h (5.0 mL/min)
Binding buffer:	50 mM MES, pH 6.0
Elution buffer:	50 mM MES, 1 M NaCl, pH 6.0
Sample:	1.5 mg/mL concanavalin A,
	1.5 mg/mL ribonuclease A,
	0.5 mg/mL a-chymotrypsinogen A,
	0.5 mg/mL lysozyme in binding buffer
Sample volume:	4.6 mL
Linear gradient	0-50% elution buffer in 20 CV



Separation using cation exchange chromatography. Peaks from left to right, concanavalin A,  $\alpha$ -chymotrypsinogen A, ribonuclease A and lysozyme. 4.6 mL sample applied on GoBio Prep 16x100 40S. The blue trace corresponds to absorbance at 280 nm and the red line to conductivity.

# Peptide purification, comparison of dynamic binding capacity at different flow rates

Column:
Sample:
Binding buffer:
Elution buffer:
Flow rates:
Residence times:

 $\begin{array}{l} GoBio \, Mini\,S\,1\,mL \\ 1\,mg\,39\,aa-long\,peptide\,(pure) \\ 5\,mM\,NH_4Ac, 15\%\,ACN, pH\,4.1 \\ 250\,mM\,NH_4Ac, 15\%\,ACN, pH\,5.6 \\ 0.3, 0.6, 1\,and\,2\,mL/min \\ 4, 2, 1\,and\,0.5\,minutes \end{array}$ 



Relationship between DBC and residence time for a 39 aa-long peptide on GoBio Mini S (cation exchanger).

### **Technical specifications**

	WorkBeads 40S	WorkBeads 100S
Matrix	Rigid, highly cross-linked agarose	Rigid, highly cross-linked agarose
Average particle size <sup>1</sup> ( $D_{v50}$ )	45 µm	90 to 110 µm
lonic group (ligand)	Sulfonate (-SO <sub>3</sub> <sup>-</sup> )	Sulfonate (-SO <sub>3</sub> <sup>-</sup> )
lonic capacity	180 to 250 µmol H*/mL resin	180 to 250 µmol H*/mL resin
Dynamic binding capacity <sup>2</sup> (DBC)	130 mg BSA/mL resin	> 100 mg BSA/mL resin
Pressure flow characteristic	N/A	2 bar at 900 cm/h, 25 mm diameter $\times$ 20 cm bed height
Maximum flow rate	600 cm/h (20 cm bed height, 5 bar)	N/A
Chemical stability	Compatible with all standard aqueous buffers used for protein purification, 1 M NaOH, 30% isopropanol or 70% ethanol. Should not be stored at < pH 3 for prolonged time.	
Operational pH range <sup>3</sup>	3 to 12	3 to 12
CIP and screening pH range <sup>3</sup>	2 to 14	2 to 14
Storage	$2\text{to}25^\circ\text{C}$ in 20% ethanol with 0.2 M sodium acetate	$2\text{to}25^\circ\text{C}\text{in}20\%\text{ethanol}$ with $0.2M\text{sodium}\text{acetate}$

The median particle size of the cumulative volume distribution.

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Dynamic binding capacity determined at 4 minutes residence time in 20 mM Na-citrate, pH 4.0. Within the operational pH range, the resin can be operated without significant change in function. Within the CIP (Cleaning-in-place) and screening pH range the resin can be subjected to the з denoted pH range without significant change in function

## Ordering information

Product name	Pack size	Article number
WorkBeads 40S	25 mL 200 mL 1 L 5 L 10 L	40 200 001 40 200 002 40 200 010 40 200 050 40 200 060
WorkBeads 100S	25 mL 200 mL 1L 5 L 10 L	10 200 001 10 200 002 10 200 010 10 200 050 10 200 060



#### Data Sheet, DS 40 100 010

WorkBeads 40S, WorkBeads 40Q, WorkBeads 40 DEAE

GoBio Mini IEX Screening kit, GoBio Mini Peptide Purification kit, GoBio prepacked columns

#### Data Sheet, DS 10 200 010

WorkBeads 100S, WorkBeads 100Q GoBio prepacked columns

→ bio-works.com/product/iex-resin



### Anion exchangers



### WorkBeads 40Q WorkBeads 100Q WorkBeads 40 DEAE

- Available with two different ligands, strong anion exchanger (Q) and weak anion exchanger (DEAE)
- Designed for research and industrial scale purifications of proteins, peptides and oligonucleotides
- Two different bead sizes, 45 μm and 100 μm
- · High chemical stability for easy cleaning-in-place
- · High binding capacity also during high flow rates
- Available in several different GoBio prepacked columns



Structure of the ligand used in WorkBeads 40Q



### Application

#### Separation on GoBio Prep 16x100 40Q

Column:	GoBio Prep 16x100 40Q (20 mL)
Flow rate:	150  cm/h (5.0  mL/min)
Binding buffer:	50 mM Tris-HCl, pH 7.4
Elution buffer:	50 mM Tris-HCl, 1M NaCl, pH 7.4
Sample:	0.3 mg/mL apo-transferrin,
	0.2 mg/mL α-lactalbumin,
	0.6 mg/mL soybean trypsin inhibitor in binding buffer
Sample volume:	46 mL
Linear gradient:	0 – 40% elution buffer in 20 CV



Separation using anion exchange chromatography. Peaks from left to right, apo-transferrin,  $\alpha$ -lactalbumin and soybean trypsin inhibitor. 46 mL sample applied on GoBio Prep 16x100 40Q. The blue trace corresponds to absorbance at 280 nm and the red line to conductivity.



### **Technical specifications**

	WorkBeads 40Q	WorkBeads 100Q
Matrix	Rigid, highly cross-linked agarose	Rigid, highly cross-linked agarose
Average particle size <sup>1</sup> $(D_{v50})$	45 µm	90 to 110 µm
lonic group (ligand)	Quaternary amine (-N⁺(CH₃)₃)	Quaternary amine (-N⁺(CH₃)₃)
lonic capacity	180 to 250 µmol Cl <sup>-</sup> /mL resin	140 to 200 µmol Cl <sup>-</sup> /mL resin
Dynamic binding capacity <sup>2</sup> (DBC)	47 mg BSA/mL resin	> 40 mg BSA/mL resin
Pressure flow characteristic	N/A	2 bar at 900 cm/h, 25 mm diameter × 20 cm bed height
Maximum flow rate <sup>3</sup>	600 cm/h (20 cm bed height, 5 bar)	N/A
Chemical stability	Compatible with all standard aqueous buffers used for protein purification, 1M NaOH, 30% isopropanol or 70% ethanol. Should not be stored at < pH 3 for prolonged time.	
Operational pH range <sup>4</sup>	2 to 13	2 to 13
CIP and screening pH range <sup>4</sup>	2 to 14	2 to 14
Storage	2 to 25°C in 20% ethanol	2 to 25°C in 20% ethanol

The median particle size of the cumulative volume distribution. Dynamic binding capacity determined at 2.5 minutes residence time in 50 mM Tris-HCl, 50 mM NaCl, pH 8.0. Maximum flow rate for aqueous buffers at 20°C. Decrease the maximum flow rate if the liquid has a higher viscosity. Higher viscosities can be caused by low temperature (use half of the

maximum flow rate for 20% ethanol). Within the operational pH range, the resin can be operated without significant change in function. Within the CIP (Cleaning-in-place) and screening pH range the resin can be subjected to the 4 denoted pH range without significant change in function.

	WorkBeads 40 DEAE
Matrix	Rigid, highly cross-linked agarose
Average particle size <sup>1</sup> $(D_{v50})$	45 µm
lonic group (ligand)	Diethylaminoethyl (-CH <sub>2</sub> CH <sub>2</sub> N*H(CH <sub>2</sub> CH <sub>3</sub> ) <sub>2</sub> )
lon capacity	110 to 160 µmol Cl <sup>-</sup> /mL resin
Dynamic binding capacity <sup>2</sup> (DBC)	40 mg BSA/mL resin
Maximum flow rate <sup>3</sup>	600 cm/h (20 cm bed height, 5 bar)
Chemical stability	Compatible with all standard aqueous buffers used for protein purification, 1 M NaOH, 30% isopropanol or 70% ethanol. Should not be stored at < pH 3 for prolonged time
Operational pH range <sup>₄</sup>	2 to 13
CIP and screening pH range⁴	2 to 14
Storage	2 to 25°C in 20% ethanol

The median particle size of the cumulative volume distribution. Dynamic binding capacity determined at 4 minutes residence time (0.25 mL/min in 1 mL column) in 50 mM Tris-HCl, pH 8.0. Maximum flow rate for aqueous buffers at 20°C. Decrease the maximum flow rate if the liquid has a higher viscosity. Higher viscosities can be caused by low temperature (use half of the maximum flow rate for 20% ethanol). 3

Within the operational pH range, the resin can be operated without significant change in function. Within the CIP (Cleaning-in-place) and screening pH range the resin can be subjected to the denoted pH range without significant change in function.

# Ordering information

Product name	Pack size	Article number
WorkBeads 40Q	25 mL 200 mL 1 L 5 L 10 L	40 100 001 40 100 002 40 100 010 40 100 050 40 100 060
WorkBeads 100Q	25 mL 200 mL 1 L 5 L 10 L	10 100 001 10 100 002 10 100 010 10 100 050 10 100 060
WorkBeads 40 DEAE	25 mL 200 mL 1L 5 L 10 L	40 150 001 40 150 002 40 150 010 40 150 050 40 150 060



#### Data Sheet, DS 40 100 010

WorkBeads 40S, WorkBeads 40Q, WorkBeads 40 DEAE GoBio Mini IEX Screening kit, GoBio Mini Peptide Purification kit, GoBio prepacked columns

#### Data Sheet, DS 10 200 010

WorkBeads 100S, WorkBeads 100Q, GoBio prepacked columns

### → bio-works.com/product/iex-resin



