WorkBeads 40S
WorkBeads 40Q

The WorkBeads™ 40S and WorkBeads 40Q resins for ion exchange chromatography are designed for research and industrial scale purification of proteins, peptides and oligonucleotides by utilizing the difference in their surface charge. WorkBeads 40S resin is a strong cation exchanger derivatized with sulfonate ligands, and WorkBeads 40Q resin is a strong anion exchanger derivatized with quaternary amine ligands. These resins demonstrate the property of high-resolution separation while giving low backpressure facilitate both capture and polishing purification applications in standard bioprocess columns.

- High throughput and purity
- Reliable and reproducible results
- High chemical stability for easy cleaning-in-place

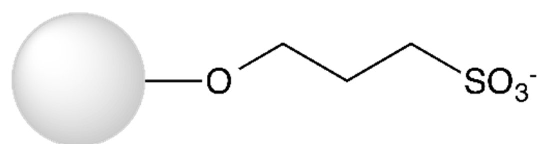
Resin description

WorkBeads are agarose-based chromatographic resins manufactured using a proprietary method that results in porous beads with a tight size distribution and exceptional mechanical stability. Agarose based matrices have been successfully used for decades in biotechnology purification, from research to production scale, due to their exceptional compatibility with biomolecules including proteins, peptides, nucleic acids and carbohydrates. WorkBeads resins are designed for separations requiring optimal capacity and purity.
WorkBeads 40S is a strong cation exchange resin derivatized with sulfonates as functional groups. WorkBeads 40Q is a strong anion exchanger derivatized with quarternary amines as functional groups.

The functional groups are coupled to the resin via chemically stable linkages. The structures of the ligands used in WorkBeads 40S and WorkBeads 40Q are shown in Figure 1.

The main characteristics of WorkBeads 40S and WorkBeads 40Q resins are shown in Table 1. For more details, please see IN 40 100 010.

(A)



(B)

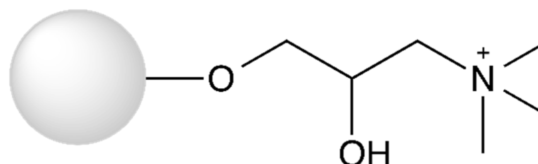


Figure 1. Structure of the ligand used in (A) WorkBeads 40S and (B) WorkBeads 40Q.

Table 1. Main characteristics of the WorkBeads 40S and WorkBeads 40Q resins.

	WorkBeads 40S	**WorkBeads 40Q**
Target substances	Proteins and peptides	Proteins, peptides and oligonucleotides
Matrix	Rigid, highly cross-linked agarose	Rigid, highly cross-linked agarose
Average particle size[1] (D_{v50})	45 µm	45 µm
Ionic group (ligand)	Sulfonate ($-SO_3^-$)	Quarternary amine ($-N^+(CH_3)_3$)
Ionic capacity	180 - 250 µmol H^+/ml resin	180 - 250 µmol Cl^-/ml resin
Dynamic binding capacity (DBC)	130 mg BSA/ ml resin[2]	47 mg BSA/ml resin[3]
Max flow rate (20 cm bed height and 5 bar)	600 cm/h	600 cm/h
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	
pH stability	2 - 13	2 - 13
Storage	2 to 25°C in 20% ethanol with 0.2 M sodium acetate	2 to 25°C in 20% ethanol

1. The median particle size of the cumulative volume distribution.
2. Dynamic binding capacity determined at 4-minutes residence time in 20 mM Na-citrate, pH 4.0.
3. Dynamic binding capacity determined at 2.5-minutes residence time in 50 mM Tris-HCl, 50 mM NaCl, pH 8.0.

Applications

WorkBeads 40S and WorkBeads 40Q resins are designed for ion exchange chromatography (IEX). The resins can be used for research- and industrial-scale purification of proteins, peptides and oligonucleotides. The particle size has been selected to enable high-resolution separations at moderate backpressure. The resins can therefore be used in capture step purification as well as for polishing. For purification requiring exceptionally high flow rates in order to handle large sample volumes WorkBeads 100S and WorkBeads 100Q should be considered for the capture step.

Principle

Ion exchange chromatography separates biomolecules according to surface charge. For example, protein interact with different affinities with opposite charged groups on the resin that depend both on the number of charges involved in the interaction and on the distribution of the charges on the protein. The surface charge of proteins depends on the pH of their environment. When the pH is equal to the isoelectric point (pI) of the protein the net charge is zero. At pH values below the pI the net charge will be positive, and at a pH above the pI the net charge will be negative. It should be noted that the interaction of the protein depends on the presence and distribution of both positive and negative charged groups on the surface. A protein may therefore also interact with an ion exchange resin at the isoelectric point.

The likelihood of binding to either the cation or the anion exchange resin will increase when the pH moves away from the protein pI.

IEX is one of the most frequently used chromatography techniques because of its versatility and ability to separate proteins even with small differences in charge and is a concentration step. It is one of the more cost-effective chromatography techniques and is excellent for scale-up.

For additional information about the ion exchange chromatography principle, see instruction IN 40 100 010.

Protein selectivity

Below, two examples are presented. In Figure 2, the basic proteins, concanavalin A, ribonuclease A, α-chymotrypsinogen A and lysozyme, are separated using WorkBeads 40S, packed in a glass column of 10 cm bed height. In Figure 3, the acidic proteins apo-transferrin, α-lactalbumin and soybean trypsin inhibitor are separated using WorkBeads 40Q, packed in a glass column with 100 mm bed height.

Column:	WorkBeads 40S in 6.6 × 100 mm, 3.42 ml
Binding buffer:	50 mM MES, pH 6.0
Elution buffer:	50 mM MES, 1 M NaCl, pH 6.0
Sample:	1 ml of 1.5 mg/ml of concanavalin A, 1.5 mg/ml ribonuclease A, 0.5 mg/ml of α-chymotrypsinogen A and 0.5 mg/ml lysozyme in binding buffer
Flow rate:	150 cm/h, 4-minute residence time
Gradient:	0 - 50% elution buffer in 20 CV

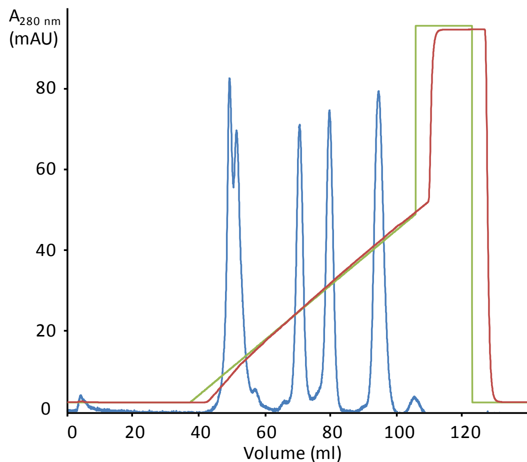


Figure 2. Separation on a cation exchange chromatography column of, peaks from left to right, concanavalin A, α-chymotrypsinogen A, ribonuclease A and lysozyme. A 1-ml sample was applied onto WorkBeads 40S column, 100 mm bed height. The blue line corresponds to the absorbance at 280 nm, the red line to the conductivity and the green line to the percentage of the elution buffer.

Column:	WorkBeads 40Q in 6.6 × 100 mm, 3.42 ml
Binding buffer:	50 mM Tris-HCl, pH 7.4
Elution buffer:	50 mM Tris-HCl, 1 M NaCl, pH 7.4
Sample:	10 ml of 0.3 mg/ml apo-transferrin, 0.2 mg/ml α-lact-albumin and 0.6 mg/ml soybean trypsin inhibitor in binding buffer
Flow rate:	150 cm/h, 4-minutes residence time
Gradient:	0 - 40% elution buffer in 20 CV

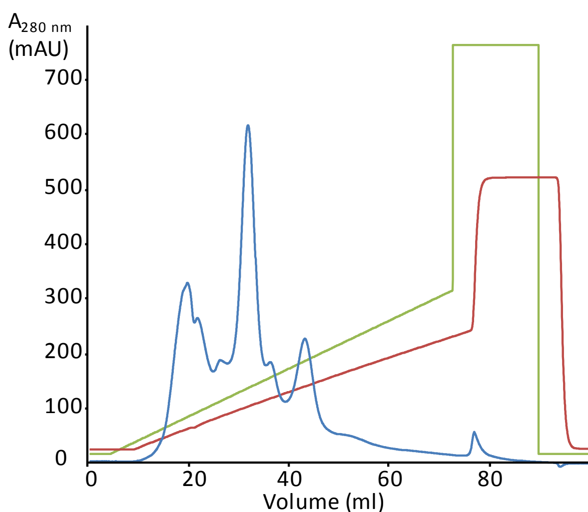


Figure 3. Separation on anion exchange chromatography column of, peaks from left to right, apo-transferrin, α-lactalbumin and soybean trypsin inhibitor. 10 ml sample applied onto a column packed with WorkBeads 40Q resin. 100 mm bed height. The blue line corresponds to the absorbance at 280 nm, the red line to the conductivity and the green line to the percentage of the elution buffer.

Flow properties

WorkBeads 40S and WorkBeads 40Q ion exchange chromatography resins are designed for high throughput protein separations under a variety of conditions. The high resolution that can be obtained at high protein loadings and high flow rates makes it ideal for process applications. Pressure/flow properties for WorkBeads 40S is shown in Figure 4. The measurements were carried out with an open bed (adaptor not pushed against the bed).

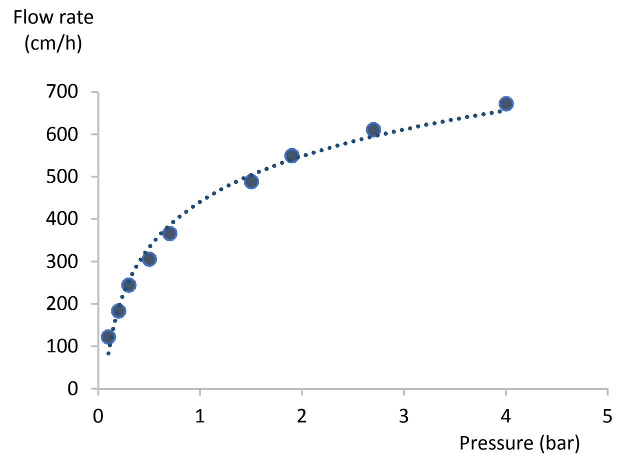


Figure 4. Pressure to flow rate properties of WorkBeads 40S determined with deionized water, 25 x 200-mm glass column.

Cleaning-in-place

During purification impurities such as cell debris, lipids, nucleic acids and protein precipitates from the samples may gradually build up in the resin bed. Fouling is typical even for well-clarified samples. The severity of this process depends on the composition of sample applied to the column. These adsorbed impurities will reduce the performance of the column over time. Regular cleaning (Cleaning-in-place, CIP) keeps the resin clean, reduces the rate of further fouling, and maintains the capacity, resolution and flow properties of the column. Cleaning of a column using 1 M NaOH using a low reversed flow for 2 hours or overnight is often sufficient.

Sanitization (reduction of microorganisms) can be carried out using combinations of NaOH and ethanol (e.g., incubation with a mixture of 0.5 M NaOH and 40% ethanol for 3 hours). The sanitization procedure and its effectiveness will depend on the microorganisms to be sanitized and needs to be evaluated for each case.

Storage

Store at 2 to 25°C in 20% ethanol. For WorkBeads 40S it is recommended to include 0.2 M sodium acetate in the storage solution.

Related products

Product name	Pack size[1]	Article number
Prepacked columns		
BabyBio S 1 ml	1 ml x 1	45 200 101
BabyBio Q 1 ml	1 ml x 1	45 100 101
BabyBio DEAE 1 ml	1 ml x 1	45 150 101
BabyBio Dsalt 5 ml	5 ml x 1	45 360 105
OptioBio 40S 10x100	7.9 ml x 1	55 420 011
OptioBio 40Q 10x100	7.9 ml x 1	55 410 011
Bulk resins		
WorkBeads 40 DEAE	25 ml	40 150 001
WorkBeads 100S	25 ml	10 200 001
	200 ml	10 200 002
	500 ml	10 200 005
	1 L	10 200 010
	5 L	10 200 050
	10 L	10 200 060
WorkBeads 100Q	25 ml	10 210 001
	200 ml	10 210 002
	500 ml	10 210 005
	1 L	10 210 010
	5 L	10 210 050
	10 L	10 210 060

1. Other pack sizes can be found in the complete product list on www.bio-works.com

Ordering information

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

Orders: sales@bio-works.com or contact your local distributor.

For more information about local distributor and products please visit www.bio-works.com or contact us at info@bio-works.com

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