

## OptioBio 40S 10x100

## OptioBio 40Q 10x100

The prepacked OptioBio™ glass columns are designed for small-scale purification as well as screening and optimization in bioprocess development and scale-up. OptioBio 40S 10x100 and OptioBio 40Q 10x100 columns are prepacked with WorkBeads™ 40S and WorkBeads 40Q resins for ion exchange chromatography (IEC). The resins are designed for research and industrial scale purification of proteins, peptides and nucleic acids and utilise the difference in their surface charge. WorkBeads 40S is a strong cation exchange resin with sulfonate ligands, and WorkBeads 40Q is a strong anion exchange resin with quaternary amine ligands. The property of high-resolution separation in combination with low backpressure facilitates both capture and polishing purification applications.

- Prepacked for reliable and reproducible results
- Optimal for high-performance small-scale purification and method optimization in bioprocess development
- High throughput and purity



### Resin description

WorkBeads are agarose based chromatographic resins manufactured using a proprietary method that results in porous beads with a tight size distribution and high mechanical stability. Agarose based matrices have been successfully used for decades in biotechnology, from research to production scale purifications, due to their exceptional compatibility with biomolecules including proteins, peptides, nucleic acids and carbohydrates. WorkBeads resins are designed for separations that require 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 quaternary 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.

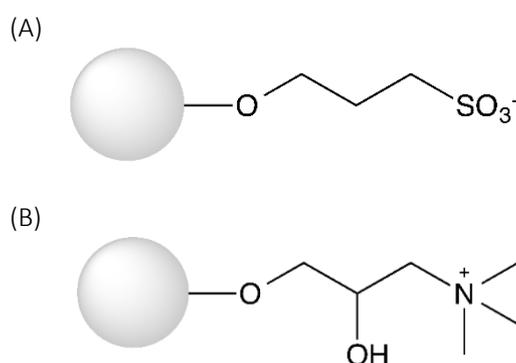


Figure 1. Structure of the ligand used in (A) WorkBeads 40S and (B) WorkBeads 40Q.

The main characteristics of OptioBio prepacked glass columns for ion exchange chromatography are shown in Table 1. For more details, please see instruction IN 55 410 010.

Table 1. Main characteristics of OptioBio 40S 10x100 and OptioBio 40Q 10x100 columns.

	OptioBio 40S 10x100	OptioBio 40Q 10x100
Target substance	Proteins and peptides	Protein, peptides and oligonucleotides
Resin	WorkBeads 40S	WorkBeads 40Q
Matrix	Rigid, highly cross-linked agarose	Rigid, highly cross-linked agarose
Average particle size (DV <sub>50</sub> ) <sup>1</sup>	45 µm	45 µm
Ionic group (ligand)	Sulfonate (-SO <sub>3</sub> <sup>-</sup> )	Quarternary amine (-N <sup>+</sup> (CH <sub>3</sub> ) <sub>3</sub> )
Ionic capacity	0.18 - 0.25 mmol Na <sup>+</sup> /ml resin	0.18 - 0.25 mmol Cl <sup>-</sup> /ml resin
Dynamic binding capacity (DBC)	150 mg BSA/ml resin <sup>2</sup>	47 mg BSA/ml resin <sup>3</sup>
Column volume (CV)	7.9 ml	7.9 ml
Column dimension	10 x 100 mm	10 x 100 mm
Recommended flow rate	2 - 4 ml/min (150 - 300 cm/h)	2 - 4 ml/min (150 - 300 cm/h)
Maximum flow rate <sup>4</sup>	6 ml/min (450 cm/h)	6 ml/min (450 cm/h)
Column hardware pressure limit	2.1 MPa, 21 bar, 305 psi	2.1 MPa, 21 bar, 305 psi
Chemical stability	Compatible with all standard 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	3 - 12 (working range) 2 - 13 (cleaning)	3 - 12 (working range) 2 - 13 (cleaning)
Storage	2 to 25 °C in 20% ethanol	2 to 25 °C in 20% ethanol

1. The median particle size of the cumulative volume distribution.

2. Dynamic binding capacity determined in 20 mM Na-citrate, pH 4.0, at a flow of 2 ml/min (150 cm/h; 4 minutes residence time).

3. Dynamic binding capacity determined in 50 mM Tris-HCl, 50 mM NaCl, pH 8.0, at a flow of 2 ml/min (150 cm/h; 4 minutes residence time).

4. 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.

## Column description

The column is made from borosilicate glass. The top and bottom filters are polyethylene and the adaptors are PEEK with 10/32 female connection for 1/16" tubing.

The ready-to-use glass columns are delivered with plugs in the inlet and outlet for storage. These columns can easily be connected to a pump or chromatography system using fingertight fittings (coned 10/32) for 1/16" o.d. tubing (standard HPLC PEEK tubing). The design of the columns, including the adaptors, have low dead volume and therefore low band broadening.

The prepacked OptioBio columns for ion exchange chromatography are designed for small scale purification as well as screening and optimization in bioprocess development and scale-up. The columns can be used for easy and reproducible scale-up purification after screening or small scale purification on prepacked BabyBio™ columns. Alternatively used directly for screening and optimization in bioprocess development.

## Applications

### IEC principle

Ion exchange chromatography separates biomolecules according to surface charge. Proteins interact with different affinities with opposite charged groups on the resin.

The interaction depends both on the number of charges involved 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.

IEC is one of the most frequently used chromatography techniques because of its versatility and ability to separate proteins even with small differences in charge. This feature, in combination with that IEC in binding mode, also works as a concentration step makes it a very efficient tool. IEC is one of the more cost-effective chromatography techniques and it is excellent for scale-up.

For additional information about the ion exchange chromatography principle, see instruction IN 55 410 010.

## Protein selectivity

In Figure 2, a set of basic proteins are separated on OptioBio 40S 10x100. In Figure 3, a set of acidic proteins are separated using OptioBio 40Q 10x100.

Column: OptioBio 40S 10 x 100  
 Binding buffer: 50 mM MES, pH 6.0  
 Elution buffer: 50 mM MES, 1 M NaCl, pH 6.0  
 Sample: 2.5 ml 1.5 mg/ml of Concanavalin A, 0.5 mg/ml  $\alpha$ -Chymotrypsinogen A, 1.5 mg/ml Ribonuclease A, 0.5 mg/ml Lysozyme in binding buffer  
 Flow rate: 2 ml/min, 150 cm/h, 4 minutes residence time  
 Gradient: 0 - 50% elution buffer in 20 column volumes (CV)

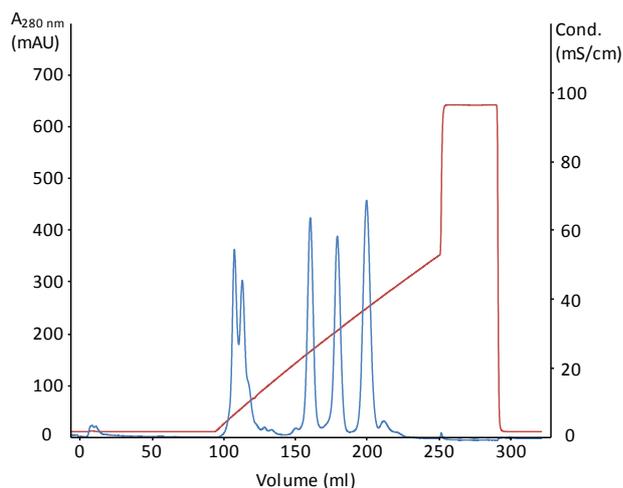


Figure 2. Separation on OptioBio 40S 10x100 prepacked strong cation exchange chromatography column. Peaks from left to right, Concanavalin A,  $\alpha$ -Chymotrypsinogen A, Ribonuclease and Lysozyme. The blue line corresponds to the absorbance at 280 nm and the red line to the conductivity.

Column: OptioBio 40Q 10 x 100  
 Binding buffer: 50 mM Tris-HCl, pH 7.4  
 Elution buffer: 50 mM Tris-HCl, 1 M NaCl, pH 7.4  
 Sample: 10 ml 0.7 mg/ml apo-Transferrin, 0.45 mg/ml  $\alpha$ -Lactalbumin, 1.4 mg/ml Soybean trypsin inhibitor in binding buffer  
 Flow rate: 2 ml/min, 150 cm/h, 4 minutes residence time  
 Gradient: 0 - 40% elution buffer in 20 CV

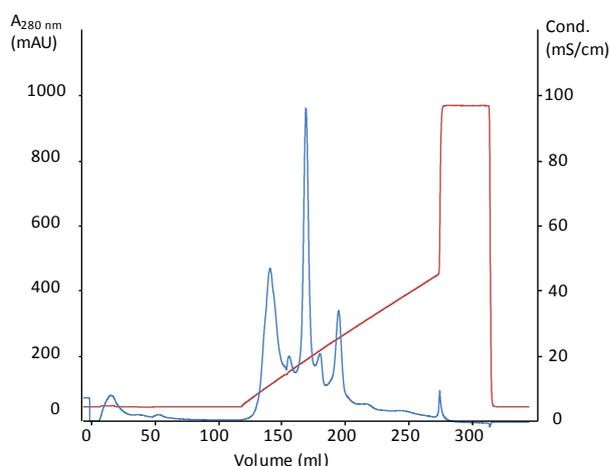


Figure 3. Separation on OptioBio 40Q 10x100 prepacked strong anion exchange chromatography column. Peaks from left to right, apo-Transferrin,  $\alpha$ -Lactalbumin and Soybean trypsin inhibitor. The blue line corresponds to the absorbance at 280 nm and the red line to the conductivity.

## Flow properties

WorkBeads 40S and WorkBeads 40Q ion exchange chromatography resins are designed for high throughput protein separation under various conditions. The high resolution obtainable even at high protein loadings and high flows makes it ideal for process applications when large volumes are processed.

In Figure 4, the operating pressure-flow curve for OptioBio 40S 10x100 column is shown in comparison with WorkBeads 40S pressure-flow curve in a 25x200 mm glass column with open bed (adaptor not pushed against the bed).

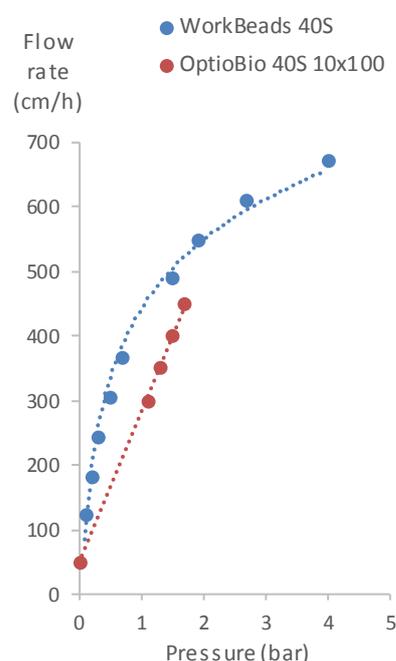


Figure 4. Pressure-flow properties of WorkBeads 40S determined with deionized water. Blue dots open bed in 25x200 mm glass column, red dots operating pressure-flow properties for OptioBio 40S 10x100.

## Cleaning and sanitization

During purification, impurities such as cell debris, lipids, nucleic acids and protein precipitates from the samples may gradually build up in the resin. Fouling is typical even for well-clarified samples. The severity of this process depends on the composition of the sample applied to the column. These adsorbed impurities will reduce the performance of the packed 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 packed column. Cleaning of the packed column using 1 M NaOH applied by a low flow for 2 hours or overnight is often sufficient. If possible, perform the CIP using reversed flow to release any particles derived from the sample that may have collected on the top filter.

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 removed, and needs to be evaluated for each case.

## Equipment

The prepacked OptioBio glass columns can be used with most standard liquid chromatography equipment.

## Storage

Store the prepacked column at 2 to 25°C in 20 % ethanol. Make sure that the column is securely closed.

## Related products

Related product	Pack size <sup>1</sup>	Article number
<b>Prepacked columns</b>		
BabyBio S 1 ml	1	45 200 101
BabyBio Q 1 ml	1	45 100 101
BabyBio DEAE 1 ml	1	45 150 101
BabyBio Dsalt 5 ml	1	45 360 105
<b>Bulk resins</b>		
WorkBeads 40S	25 ml	40 200 001
	200 ml	40 200 002
	1 L	40 200 010
	5 L	40 200 050
WorkBeads 40Q	25 ml	40 100 001
	200 ml	40 100 002
	1 L	40 100 010
	5 L	40 100 050
WorkBeads 40 DEAE	25 ml	40 150 001
WorkBeads 100S	25 ml	10 200 001
WorkBeads 100Q	25 ml	10 210 001

1. Other pack sizes can be found in the complete product list on our website [www.bio-works.com](http://www.bio-works.com).

## Ordering information

Product name	Pack size	Article number
OptioBio 40S 10x100	7.9 ml	55 420 011
OptioBio 40Q 10x100	7.9 ml	55 410 011

Orders: [sales@bio-works.com](mailto:sales@bio-works.com) or contact your local distributor.

For more information about local distributor and products, please visit [www.bio-works.com](http://www.bio-works.com) or contact us at [info@bio-works.com](mailto:info@bio-works.com)



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