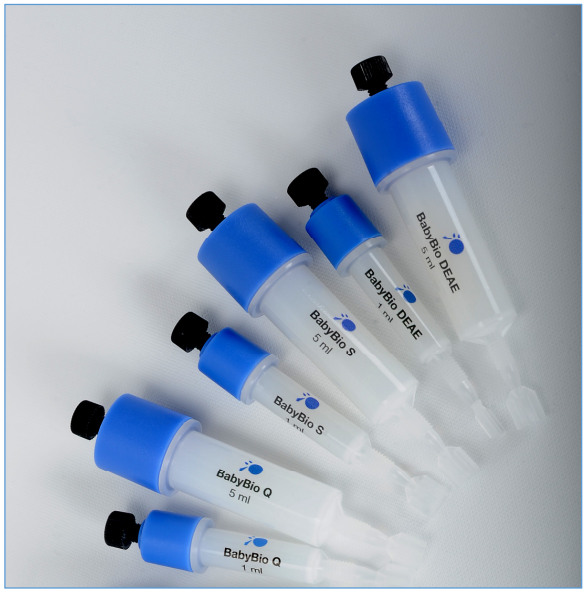
BabyBio S
BabyBio Q
BabyBio DEAE

BabyBio™ S, BabyBio Q and BabyBio DEAE are ready-to-use ion exchange chromatography columns for easy and convenient purification of proteins, peptides and oligonucleotides by utilizing the difference in the surface charge of the target molecule BabyBio S works as a strong cation exchanger, BabyBio Q as a strong anion exchanger and BabyBio DEAE as a weak anion exchanger. The columns are prepacked with WorkBeads™ 40S, WorkBeads 40Q and WorkBeads 40 DEAE resins, and are available in two sizes, 1 ml and 5 ml.

- Prepacked, ready-to-use, for fast and reliable results
- High binding capacity and purity
- Easy-to-use for the screening of conditions



Resin description

BabyBio S, BabyBio Q and BabyBio DEAE are prepacked with resins manufactured using a proprietary method that results in porous beads with a tight size distribution and exceptional mechanical stability. WorkBeads resins are designed for separations that require optimal capacity and purity.

BabyBio S columns are packed with WorkBeads 40S resin, a strong cation exchanger with sulfonate functional groups.

BabyBio Q columns are packed with WorkBeads 40Q resin, a strong anion exchanger with quarternary amine functional groups. BabyBio DEAE columns are packed with WorkBeads 40 DEAE resin, a weak anion exchanger with tertiary amine functional groups.
The functional groups are coupled to the resin via chemically stable linkages. The structures of the ligands used in WorkBeads 40S, WorkBeads 40Q and WorkBeads 40 DEAE are shown in Figure 1.

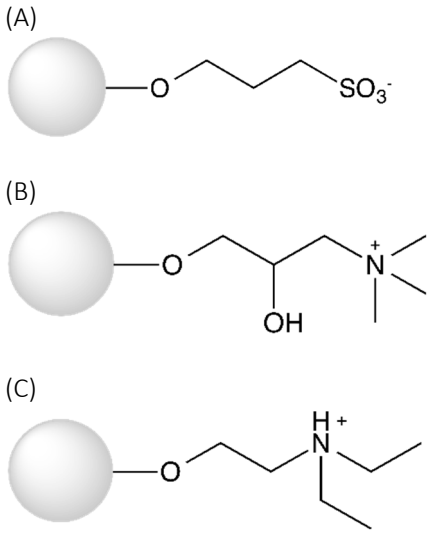


Figure 1. Structure of the ligand used in (A) WorkBeads 40S, (B) WorkBeads 40Q and (C) WorkBeads 40 DEAE.

The main characteristics of BabyBio S, BabyBio Q and BabyBio DEAE are shown in Table 1. For more details, please see instruction, IN 45 100 010.

Table 1. Main characteristics of BabyBio S, BabyBio Q and BabyBio DEAE 1 ml and 5 ml columns.

	BabyBio S	BabyBio Q	BabyBio DEAE
Target substance	Proteins, peptides	Protein, peptides, oligonucleotides	Protein, peptides, oligonucleotides
Resin	WorkBeads 40S	WorkBeads 40Q	WorkBeads 40 DEAE
Matrix	Rigid, highly cross-linked agarose	Rigid, highly cross-linked agarose	Rigid, highly cross-linked agarose
Average particle size[1] (D_{V50})	45 µm	45 µm	45 µm
Ligand	Sulfonate ($-SO_3^-$)	Quarternary amine ($-N^+(CH_3)_3$)	Diethylaminoethyl ($-CH_2CH_2N^+H(CH_2CH_3)_2$)
Ion capacity	0.18 - 0.25 mmol Na^+/ml resin	0.18 - 0.25 mmol Cl^-/ml resin	0.11 - 0.16 mmol Cl^-/ml resin
Dynamic binding capacity	130 mg BSA/ml resin[2]	50 mg BSA/ml resin[3]	40 mg BSA/ml resin[3]
Column volume	1 ml 5 ml	1 ml 5 ml	1 ml 5 ml
Column dimension	7 x 28 mm (1 ml) 13 x 38 mm (5 ml)	7 x 28 mm (1 ml) 13 x 38 mm (5 ml)	7 x 28 mm (1 ml) 13 x 38 mm (5 ml)
Recommended flow rate			
BabyBio 1 ml	1 ml/min (150 cm/h)	1 ml/min (150 cm/h)	1 ml/min (150 cm/h)
BabyBio 5 ml	5 ml/min (225 cm/h)	5 ml/min (225 cm/h)	5 ml/min (225 cm/h)
Maximum flow rate			
BabyBio 1 ml	5 ml/min (780 cm/h)	5 ml/min (780 cm/h)	5 ml/min (780 cm/h)
BabyBio 5 ml	20 ml/min (900 cm/h)	20 ml/min (900 cm/h)	20 ml/min (900 cm/h)
Maximum back pressure	0.3 MPa, 3 bar, 43 psi	0.3 MPa, 3 bar, 43 psi	0.3 MPa, 3 bar, 43 psi
Chemical stability	Compatible with all standard aqueous buffers used for protein purification and 70% ethanol. Should not be stored at low pH for prolonged time		
pH stability	2 - 13	2 - 13	3 - 9 recommended pH 3 - 13
Storage	2 to 25 °C in 20% ethanol	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 at 4 minutes residence time (0.25 ml/min in 1 ml column) in 20 mM Na-citrate, 60 mM NaCl, pH 4.0.
3. Dynamic binding capacity determined at 4 minutes residence time (0.25 ml/min in 1 ml column) in 50 mM Tris-HCl, 50 mM NaCl, pH 8.0.
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 biocompatible polypropylene which does not significantly interact with biomolecules. The top and bottom filters are made from polyethylene. The ready-to-use BabyBio columns are delivered with a plug in the inlet, a cut-off outlet and a cap for storage. The columns can be connected to a syringe, pump or chromatography system using finger tight fittings (coned 10–32) for 1/16” o.d. tubing (standard HPLC PEEK tubing).

Applications

These BabyBio columns are designed for Ion Exchange Chromatography (IEX). BabyBio S, BabyBio Q and BabyBio DEAE columns can successfully be used in the separation of proteins or other biomolecules in biotechnology research. The resin are designed to maintain high capacity over broad pH ranges.

Principle

Ion exchange chromatography separates biomolecules according to surface charge. For example, proteins interact with different affinities with opposite charged groups on the resin. This depends 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, the net charge.

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 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 also one of the more cost effective chromatography techniques and is therefore excellent for scale-up.

For additional information about the ion exchange chromatography principle, see instruction IN 45 100 010.

Protein selectivity

Below, three examples are presented. In Figure 2, the basic proteins, Concanavalin A, Ribonuclease A, α-Chymotrypsinogen A and Lysozyme are separated on BabyBio S columns. In Figure 3 and Figure 4, the acidic proteins apo-Transferrin, α-Lactalbumin and Soybean trypsin inhibitor are separated on BabyBio Q and BabyBio DEAE columns respectively.

Column:	(A) BabyBio S 1 ml, (B) BabyBio S 5 ml
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 α-Chymotrypsinogen A and 0.5 mg/ml Lysozyme in binding buffer
Flow rate:	150 cm/h (1 ml/min BabyBio S 1 ml) (3.5 ml/min BabyBio S 5 ml)
Gradient:	0 - 50% elution buffer in 20 column volumes (CV)

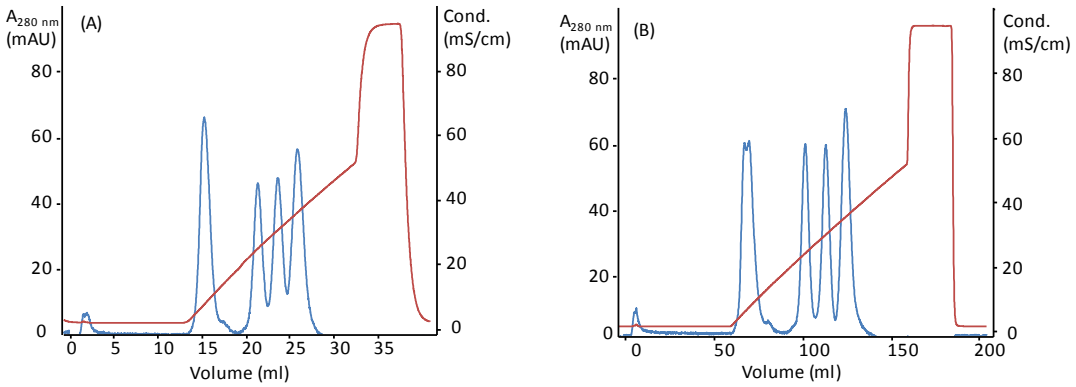


Figure 2. Separation using cation exchange chromatography. Peaks from left to right, Concanavalin A, α-Chymotrypsinogen A, Ribonuclease A and Lysozyme. (A) 0.25 ml sample applied onto a BabyBio S 1 ml and (B) 1.25 ml sample applied onto a BabyBio S 5 ml. The blue line corresponds to absorbance at 280 nm and the red line to conductivity.

Column:	(A) BabyBio Q 1 ml, (B) BabyBio Q 5 ml
Binding buffer:	50 mM Tris-HCl, pH 7.4
Elution buffer:	50 mM Tris-HCl, 1 M NaCl, pH 7.4
Sample:	0.3 mg/ml apo-Transferrin, 0.2 mg/ml α-Lactalbumin and 0.6 mg/ml Soybean trypsin inhibitor in binding buffer
Flow rate:	150 cm/h (1 ml/min BabyBio Q 1 ml) (3.5 ml/min BabyBio Q 5 ml)
Gradient:	0 - 40% elution buffer in 20 CV

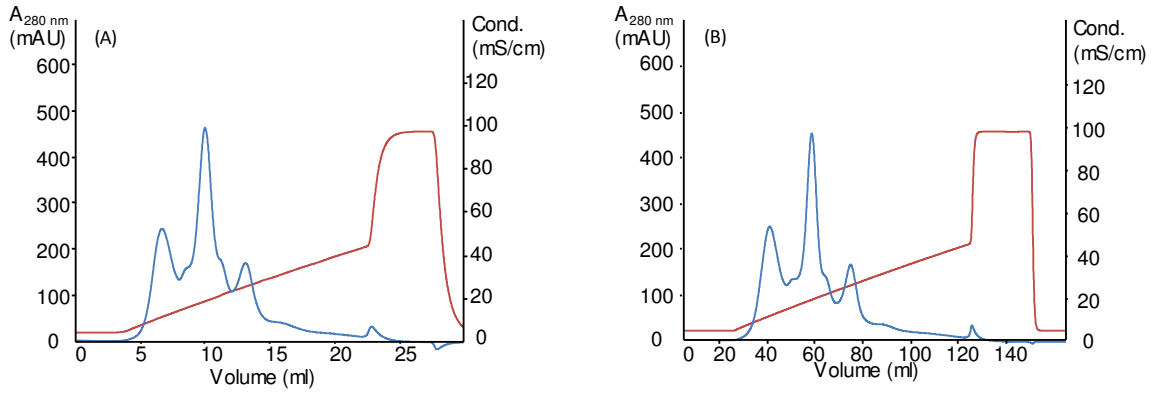


Figure 3. Separation using anion exchange chromatography. Peaks from left to right, apo-Transferrin, α-Lactalbumin and Soybean trypsin inhibitor. (A) 2 ml sample applied onto a BabyBio Q 1 ml and (B) 10 ml sample applied onto a BabyBio Q 5 ml. The blue line corresponds to absorbance at 280 nm and the red line to conductivity.

Column:	(A) BabyBio DEAE 1 ml, (B) BabyBio DEAE 5 ml
Binding buffer:	50 mM Tris-HCl, pH 7.4
Elution buffer:	50 mM Tris-HCl, 1 M NaCl, pH 7.4
Sample:	0.3 mg/ml apo-Transferrin, 0.2 mg/ml α-Lactalbumin and 0.6 mg/ml Soybean trypsin inhibitor in binding buffer
Flow rate:	150 cm/h (1 ml/min BabyBio DEAE 1 ml) (3.5 ml/min BabyBio DEAE 5 ml)
Gradient:	0 - 40% elution buffer in 20 CV

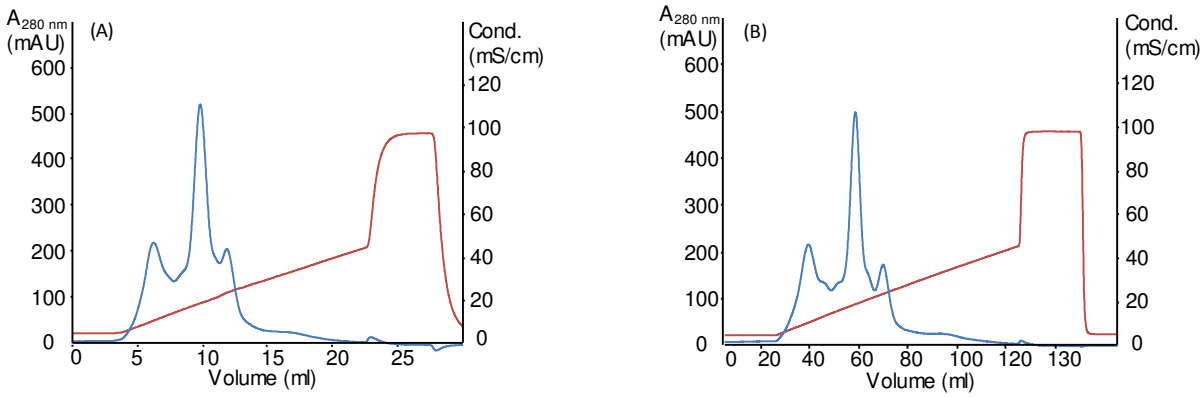


Figure 4. Separation using anion exchange chromatography. Peaks from left to right, apo-Transferrin, α-Lactalbumin and Soybean trypsin inhibitor. (A) 2 ml sample onto a BabyBio DEAE 1 ml and (B) 10 ml sample onto a BabyBio DEAE 5 ml. The blue line corresponds to absorbance at 280 nm and the red line to conductivity.

Scale-up

Scale-up can conveniently be carried out from a 1 ml column to a 5 ml column. If increased capacity is required several columns can be coupled in series (column stacking). Note that the backpressure will increase proportionally to the resin bed height (up to a maximum of 5 columns).

Further scale-up can be achieved by using prepacked OptioBio 10x100 columns (7.9 ml) or by packing bulk WorkBeads 40S, WorkBeads 40Q and WorkBeads 40 DEAE resins in larger columns (see *Related products*).

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. Fouling is typical even for well clarified samples. The severity of this process depends on the composition of sample applied to the column. The impurities adsorbed 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 column. Cleaning with 1 M NaOH applied by 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 removed, and must be evaluated for each case.

Equipment

Prepacked BabyBio S, BabyBio Q and BabyBio DEAE columns can be used with most standard liquid chromatography equipment. Purification can also be carried out using a syringe connected to the column by a luer or a std HPLC connector.

Storage

Equilibrate the column in 20% ethanol and close it securely using the included plug and cap. Store at 2 to 25°C.

Related products

Product name	Pack size[1]	Article number
Prepacked columns		
BabyBio Dsalt 1 ml	1 ml x 1	45 360 101
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 40S	25 ml	40 200 001
WorkBeads 40Q	25 ml	40 100 001
WorkBeads 40 DEAE	25 ml	40 150 001
Accessories		
Column plug male 1/16”	10	70 100 010
Column cap female 1/16”	10	70 100 020

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
BabyBio S 1 ml	1 ml x 1	45 200 101
	1 ml x 2	45 200 102
	1 ml x 5	45 200 103
	1 ml x 10	45 200 104
BabyBio S 5 ml	5 ml x 1	45 200 105
	5 ml x 2	45 200 106
	5 ml x 5	45 200 107
	5 ml x 10	45 200 108
BabyBio Q 1 ml	1 ml x 1	45 100 101
	1 ml x 2	45 100 102
	1 ml x 5	45 100 103
	1 ml x 10	45 100 104
BabyBio Q 5 ml	5 ml x 1	45 100 105
	5 ml x 2	45 100 106
	5 ml x 5	45 100 107
	5 ml x 10	45 100 108
BabyBio DEAE 1 ml	1 ml x 1	45 150 101
	1 ml x 2	45 150 102
	1 ml x 5	45 150 103
	1 ml x 10	45 150 104
BabyBio DEAE 5 ml	5 ml x 1	45 150 105
	5 ml x 2	45 150 106
	5 ml x 5	45 150 107
	5 ml x 10	45 150 108

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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