

LigaTrap[®] Loose Base Resin Product Instructions

Introduction:

LigaTrap Purification Resin is engineered to purify high quality antibodies from polyclonal and monoclonal sources. LigaTrap Purification Resin is recommended for purifying IgG in range of species as well as for Human IgA and Chicken IgY applications. Kappa and Lambda IgG, IgA, and IgY may be purified using this product. LigaTrap Purification Resin is capable of processing and purifying monoclonal antibodies form cell culture supernatant, ascites fluid, hybridoma, and other sources of recombinant IgG, IgA, and IgY.

Chromatographic Buffers:

Part #	Name	Composition		
BU-131-FP	LigaTrap Sample Diluent 2.0	50mg/mL Adipic Acid, 4.0M NaCl, pH 5.8		
BU-132-FP	LigaTrap Equilibration/Wash Buffer 2.0	10mg/mL Adipic Acid, 800mM NaCl, pH 5.8		
BU-123-FP	LigaTrap Elution Buffer	0.1M Sodium Acetate, pH 4.0		
BU-124-FP	LigaTrap Regeneration Buffer	0.1M Glycine, pH 2.5		
BU-125-FP	LigaTrap Neutralization Buffer	3.0M Tris-Base, pH 11.1		
BU-126-FP	LigaTrap Storage Buffer	10mM Sodium Phosphate, 0.15M NaCl, 0.05% Sodium Azide, pH 7.2		

Note: Adipic Acid is insoluble at low pH. It will solubilize as the pH increases to > 5.0.

Note: For best results, titrate LigaTrap Elution Buffer with Glacial Acetic Acid.

Note: To limit precipitation of Tris-Base, store LigaTrap Neutralization Buffer at room temperature.

Note: Equilibrate all buffers to room temperature prior to use.

Prepare Sample for Binding:

1. Add the **LigaTrap Sample Diluent 2.0 (BU-131-FP)** to the sample containing the target antibody at a ratio of 1:4

(For example: Add 2 mL LigaTrap Sample Diluent 2.0 to 8mL of sample, or 200mL to 800mL of sample, etc...)

Chromatographic Protocol:

- Pack the appropriately sized column volume (CV), for your particular application. Ensure not to exceed a maximum flow rate of 600cm/hr.
- ➤ If using a FPLC system capable of setting multiple pressure alarms, set the pre-column pressure alarm to 0.2 MPa.
- Recommended 5-10 minute residence times for the sample load and all other chromatographic steps
- 1. Equilibrate the resin with 10 CV of LigaTrap Equilibration/Wash Buffer 2.0 (BU-132-FP).
- 2. Load the prepared sample (described above).
- 3. Wash the resin with 10-15 CV of LigaTrap Equilibration/Wash Buffer 2.0 (BU-132-FP).
- 4. Elute bound antibody with 10 CV of LigaTrap Elution Buffer (BU-123-FP).
- 5. Neutralize with 15-18% v/v of LigaTrap Neutralization Buffer (BU-125-FP).
- 6. Regenerate the column with 10 CV of LigaTrap Regeneration Buffer (BU-124-FP).
- 7. Re-Equilibrate the resin with 10 CV **LigaTrap Equilibration/Wash Buffer 2.0 (BU-132-FP)** if an additional purification cycle will be done.

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8. If purification is complete, store the resin by running 5 CV of **LigaTrap Storage Buffer (BU-126-FP**) over the column and store at 2-8 °C.

Batch Method Preparation (Optional):

- 1. Transfer loose resin to a clean mixing container of adequate volume to hold resin volume plus sample volume and buffers.
- 2. Add mixture of sample with **LigaTrap Sample Diluent 2.0 (BU-131-FP)** into this container and mix gently for desired time, 1-2 hours mixing is recommended for a sample with very low immunoglobulin concentration or overnight at 4 °C.
- 3. Remove the supernatant after incubation and wash the resin with 5-10 CV of **LigaTrap Equilibration/Wash Buffer 2.0 (BU-132-FP)**, collect these washes.
- 4. Add **LigaTrap Elution Buffer (BU-123-FP)** and incubate for 5-10 minutes, collect the eluates. **Note:** This collected elution buffer has immunoglobulin, do not discard.
- 5. Neutralize eluates with 15-18% v/v LigaTrap Neutralization Buffer (BU-125-FP) and gently vortex.
- 6. Regenerate the resin with 5-10 CV of **LigaTrap Regeneration Buffer (BU-124-FP)** with 5-10 minutes mixing and collect the flow through.
- 7. If the resin will be reused, re-equilibrate with 10 CV **LigaTrap Equilibration/Wash Buffer 2.0 (BU-132-FP)**. Discard these washes.
- 8. If purification is complete, store the resin by adding 1 CV of **LigaTrap Storage Buffer (BU-126-FP**) into the resin and store at 2-8 °C.

Other LigaTrap Products:

		Part Number				
Product	Antibody	Loose Resin	Microspin Columns	Prepacked Columns	Purification	
LigaTrap Base Resin	lgG, lgA, lgY	LT-150	LT-150-MSC		Kits	
				LT-150-1x1mL	LT-150KIT	
				LT-150-3x1mL	LT-150-1mL KIT	
				LT-150-1x5mL	LT-150-5mL KIT	
LigaTran	lgM	LT-155	LT-155-MSC	LT-155-1x1mL	LT-155KIT	
LigaTrap IgM Resin				LT-155-3x1mL	LT-155-1mL KIT	
				LT-155-1x5mL	LT-155-5mL KIT	

For further product information, please visit our website at <u>LigaTrap.com</u>. For technical support and questions, email us at *info@ligatrap.com*

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