

LigaTrap® Chicken IgY Loose Resin Protocol

Introduction

ImmunoReagents is now offering LigaTrap® purification resins and kits, engineered for antibody purification. LigaTrap® Technologies has developed a novel, patented, series of affinity ligands specific for the purification of monoclonal and polyclonal immunoglobulins from various species. In addition to species-specific ligands, advantages include the ability to elute antibodies at a higher pH (pH 4.0) than protein A and G (pH \leq 3.0), thus reducing the potential for precipitation and inactivation of pH sensitive antibodies. LigaTrap® Chicken IgY Purification resin is capable of binding >15mg IgY/ml resin.

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	1.0M Tris, pH 8.5		
BU-126-FP	LigaTrap Storage Buffer	10mM Sodium Phosphate, 0.15M NaCl, 0.05% Sodium Azide, pH 7.2		

Note: Adipic Acid can be purchased from Sigma (A26357-500G)

<u>Note</u>: Adipic Acid is insoluble at low pH. It will solubilize as the pH increases to > 5.0. For example, after adding all components of LigaTrap Sample Diluent 2.0 Buffer, a 500ml batch will require ~60mL of 5N NaOH to begin dissolving the adipic acid. Continue to titrate with NaOH until a final pH of 5.8 is achieved. Then QS to the desired volume. Note: For best results, **titrate LigaTrap® Elution Buffer with Glacial Acetic Acid**

Prepare Sample For Binding:

1. Add the **LigaTrap Sample Diluent 2.0** to the sample containing IgY at a ratio of 1:4 (For example: Add 2ml LT 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.
- Recommend <u>5 minute residence</u> time for sample load and all other chromatographic steps
- 1. Equilibrate the resin with 10-20CV of LigaTrap Equilibration/Wash Buffer 2.0 (BU-132-FP)
- 2. Load the "prepared" sample (described above)
- 3. Wash the resin with 15-20CV of LigaTrap Equilibration/Wash Buffer 2.0 (BU-132-FP)
- 4. Elute bound antibody with 10CV of **LigaTrap Elution Buffer (BU-123-FP)**. Note, for higher concentration antibody elute with 5CV, but if higher yields are desired, use 10CV
- 5. Regenerate the column with 10CV of LigaTrap Regeneration Buffer (BU-124-FP)
- 6. Re-Equilibrate the resin with 10CV LigaTrap Equilibration/Wash Buffer 2.0 (BU-132-FP) if an additional purification cycle will be done.
- 7. If purification is complete, store the resin by running 5CV of LigaTrap Storage Buffer (BU-126-FP) over the column.

Other LigaTrap® Products

		Part Number		
Target Species	Isotype	Prepacked Columns	5mL Loose Resin	Purification Spin Column Kit
	IgG	LT-095-1x1ml		
		LT-095-3x1ml	LT-095	LT-095KIT
		LT-095-1x5ml		
	IgM	LT-143-1x1ml		
Human		LT-143-3x1ml	LT-143	LT-143KIT
		LT-143-1x5ml		
	IgA	LT-146-1x1ml		
		LT-146-3x1ml	LT-146	LT-146KIT
		LT-146-1x5ml		
	IgG	LT-137-1x1ml	LT-137	LT-137KIT
		LT-137-3x1ml		
Manaa		LT-137-1x5ml		
Mouse	IgM	LT-145-1x1ml	LT-145	LT-145KIT
		LT-145-3x1ml		
		LT-145-1x5ml		
	IgG	LT-138-1x1ml		
Rat		LT-138-3x1ml	LT-138	LT-138KIT
		LT-138-1x5ml		
	IgG	LT-141-1x1ml	LT-141	LT-141KIT
Sheep		LT-141-3x1ml		
*		LT-141-1x5ml		
	IgG	LT-144-1x1ml		
Llama		LT-144-3x1ml	LT-144	LT-144KIT
		LT-144-1x5ml		
	IgG	LT-136-1x1ml	LT-136	LT-136KIT
Goat		LT-136-3x1ml		
		LT-136-1x5ml		
	IgG	LT-139-1x1ml	LT-139	
Rabbit		LT-139-3x1ml		LT-139KIT
		LT-139-1x5ml		
	IgY	LT-142-1x1ml		
Chicken		LT-142-3x1ml	LT-142	LT-142KIT
		LT-142-1x5ml		

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