

LigATrap™ Human IgG Purification Kit, 10 x 0.15 mL

Part # *LT-095KIT*

Kit Contents

Part #	Item	Quantity
LT-095-SC	Microspin Columns - centrifuge columns supplied with caps and plug. Each column contains 0.15 mL LigATrap™ Human Resin in PBS buffer with 0.05% sodium azide.	10
BU-131-FP	LT Sample Diluent 2.0	15 mL
BU-132-FP	LT Equilibration/Wash Buffer 2.0	250 mL
BU-123-FP	LT Elution Buffer	125 mL
BU-124-FP	LT Regeneration Buffer	50 mL
BU-125-FP	LT Neutralization Buffer	15 mL
BU-126-FP	LT Storage Buffer	50 mL
PL-057	2 mL Collection Tubes	80

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 ≤ 3.0), thus reducing the potential for precipitation and inactivation of pH sensitive antibodies.

LigATrap™ Human IgG Resin is engineered to purify high quality human IgG antibodies from recombinant, monoclonal, and polyclonal sources. Isotypes of human IgG1, IgG2, and IgG3 will be bound with this resin. Binding capacity for total human IgG is >20mg/ml resin. The **LigATrap™ Human IgG Purification Kit** provides all the necessary reagents for fast, convenient micro-scale purification of human IgG in just 10 easy steps. Each kit contains 10 microspin columns prefilled with 0.15ml of **LigATrap™ Human IgG Resin**, buffers, and collection tubes. Each spin column can be used, regenerated, and used up to 10 times with minimal loss in binding capacity.

Additional Materials Required

- Microcentrifuge set between 2000-6000 x g
- Vortex/Mixer
- Centrifuge tubes or container for sample preparations

Antibody Purification Procedure

Sample Prep

1. In a separate tube (not supplied in kit) add 400µl of sample matrix (i.e. serum or cell culture media) containing human IgG
2. Add 100µl of **LT Sample Diluent 2.0** to the sample. Mix briefly by vortexing.

Purification

3. Equilibrate resin by adding 500µl of **LT Equilibration/Wash Buffer 2.0**. Snap the bottom plug on the Microspin column. Save the plug, as it will be needed to stopper the column. Insert the Microspin column into a supplied 2.0 mL Collection Tube. Centrifuge between 2000-6000 x g for 1 minute. Empty the buffer from Collection Tube. Repeat with an additional 500µl equilibration. Insert the bottom plug into the Microspin column.
4. Transfer 500µl of prepared sample (Step # 2) to the equilibrated column. Place screw cap on snugly. Vortex briefly for 15 seconds. Continue to mix/shake the sample and resin continuously for ~10 min.
5. Remove plug and insert the Microspin column into a Collection Tube. Centrifuge between 2000-6000 x g for 1 minute. Discard unbound material or retain for further evaluation.
6. Add 500µl of the **LT Equilibration/Wash Buffer 2.0**. Remove plug and insert Microspin column into a clean Collection Tube. Centrifuge between 2000-6000 x g for 1 minute. Discard wash. Repeat with two additional washes for a total of three washes. (3x500µl)

**IR-Tip: Collection Tubes used to collect washes can be reused.*

7. Insert the washed Microspin column into a **new, labeled** Collection Tube. Add 500µl of **LT Elution Buffer** to the column. Vortex briefly for 15 seconds. Centrifuge between 2000-6000 x g for 1 minute. Place the Microspin column into a new, labeled Collection Tube. Repeat with second 500µl elution. ***Note: The eluates contain the purified antibodies. Do not discard!**
8. Pool eluates from Step 7 and add 100µl of **LT Neutralization Buffer** to the antibody. Attach the cap. Vortex briefly. The antibody will be near neutral pH. The antibody is ready for downstream applications. **Note: There are no preservatives in the antibody. Use the antibody within one week or aliquot and store at -20° C or colder. Avoid multiple freeze thaws.**

**IR-Tip: Each Ligatrap™ resin column can be used up to 10 times without significant loss of binding capacity*

9. If the column will not be reused, it can be discarded. If column is to be reused, regenerate the column by adding 500µl of **LT Regeneration Buffer**. Vortex briefly for 15 seconds. Insert the Microspin column into a 2.0 mL Collection Tube and centrifuge between 2000-6000 x g for 1 minute.
10. To store resin add 500µl of **LT Storage Buffer**. Insert the Microspin column into a 2.0 mL Collection Tube. Centrifuge between 2000-6000 x g for 1 minute. Discard wash. Repeat with an additional two washes for a total of three washes. (3x500µl) Insert the bottom plug into the Microspin column, add 500µl of fresh **LT Storage Buffer** and store at 2-8° C.