### USER GUIDE: rProtein A, G, or A/G resin



### Introduction:

EBL AM-50 Protein A, G, and A/G are mediums developed with genetically modified proteins with high alkali-tolerance, which capture monoclonal antibodies for large-scale volumes with chromatography. The resins feature a variety that provides the most versatile combination of chromatographic features for rich yields and high-purity purification of whole IgG for mammalian serum samples.

### **Key Features:**

- The ligands with high alkali-tolerant (up to 0.1-0.5 NaOH) allow for cleaning-in-place (CIP).
- Improved product quality and economy with Up to 100 cycles of lifetime.
- High DBC reduces processing period and medium usage, with all the solutions and consumables saved undoubtedly.
- High-flow and high performance allows for rapid processing procedure.
- Suits for the range form small scale to industry-scale production.
- The ligands are recombinantly produced in *Escherichia coli* without animal material used in the whole process.
- The mono-dispersed polymer matrix with 50  $\mu$  m size features this product high flow rate, high performance and high DBC.

Specifications.	
Particle Size (μm)	50 ± 5 %
Bead Matrix	Highly cross-linked polyacrylate polymer
Ligand Coupling Method	Ероху
Ligand	highly Alkali-tolerant IG binding protein in E. coli.
Dynamic Binding Capacity (g/ml)	≈30 mg human IgG/ml medium
Max. Linear Flow Rate (cm/h)	700 cm/h
pH Stability	3-12
Pressure (MPa)	10
Cleaning-in-place (CIP) stability	0.1-0.5 M NaOH
Storage buffer	20 % ethanol
Storage temperature	+4-8 °C

### **Specifications:**

# **Recommended Procedure and Working Conditions:**

- Binding and Washing Buffer: 20 mM sodium phosphate, 150 mM NaCl, pH 7.2
- Elution Buffer: 0.1 M sodium citrate, pH 3.0 to 3.6.
- Collection Buffer: 1M Tris-HCl, pH 8.0 to 9.0.

\* When purifying mouse IgG1 on protein A media, an increased binding capacity will be achieved by 2.5 M NaCl included in the binding buffer

Procedure :

- 1. Adjust the composition and pH of the sample(s) to or near Binding Buffer. If needed, a buffer change step should be processed before application of the samples to the column
- 2. Wash 1 ml of settled resin with at least 5 volumes of Washing Buffer.
- 3. Apply a sample of antibody to the column.
- 4. Wash away unbound proteins with 5 column volumes of Washing Buffer.
- 5. Elute the sample with a linear gradient of 10 column volumes to 100% Elution Buffer.
- 6. Collect fractions into Collection Buffer. Please note that the Collection Buffer volume equals 5% of the programmed fraction volume.
- 7. Regenerate the column with 5-10 column volumes of 100% Elution Buffer.
- 8. Wash the column with 3 column volumes of Washing Buffer.
- 9. Perform CIP with 5 column volumes of 0.1-0.5 M NaOH.
- 10. Re-equilibrate the column with Binding Buffer.

# **About Elution:**

When optimizing elution conditions, determine the highest pH that allows efficient desorption of antibody from the column. This will prevent denaturing sensitive antibodies due to exposure to low pH.

Note: Step-wise elution is often preferred in large-scale applications since it allows the target monoclonal antibody to be eluted in a more concentrated form, thus decreasing buffer consumption and shortening cycle times. It might be necessary to decrease the flow rate due to the high concentrations of protein in the eluate.

# Ordering information:

CAT NO:	Product	Quantity
pct-p500115	packing colume protein A/G resin	1mlx 5
pct-p500111	packing colume protein A/G resin	1mlx 1
pct-p500151	packing colume protein A/G resin	5mlx 1
pct-p500215	packing colume protein A resin	1ml X 5
pct-p500211	packing colume protein A resin	1ml X 1
pct-p500251	packing colume protein A resin	5ml X 1
pct-p500315	packing colume protein G' resin	1ml X 5
pct-p500311	packing colume protein G' resin	1ml X 1
pct-p500351	packing colume protein G' resin	5ml X 1
pam-5001005	AM-50 protein A/G' resin	5ml
pam-5001020	AM-50 protein A/G' resin	20ml
pam-5001100	AM-50 protein A/G' resin	100ml
pam-5002005	AM-50 protein A	5ml
pam-5002020	AM-50 protein A	20ml
pam-5002100	AM-50 protein A	100ml
pam-5003005	AM-50 protein G'	5ml
pam-5003020	AM-50 protein G'	20ml
pam-5003100	AM-50 protein G'	100ml