Protein Purification

Chondrex, Inc offers a range of resins for protein purification. The following provides a summary on different protein purification mechanisms in order to help you choose the appropriate resins for your research and study purposes.

1. Ion-exchange Chromatography (1, 2)

Each protein has a net charge, called the isoelectric point (pI), determined by its primary amino acid sequences. This charge property allows proteins to selectively bind to resins with opposite charges in ion-exchange chromatography. This technique utilizes the following principle - positively charged resins (anion exchange: DEAE and Q), or negatively charged resins (cation exchange: CM). Then, alterations in pH or salt gradients facilitate the elution of proteins from the resin.

2. Affinity Chromatography (3)

Affinity chromatography is a highly effective method for the purification of specific proteins through biological interactions, leveraging the strong, yet reversible, binding between molecules. This method immobilizes one of the interacting proteins on a chromatographic resin, allowing for the selective binding and subsequent elution of the target protein by altering conditions, such as pH or salt concentration.

Protein Purification Resins

	Ion Exchange Purification	Antibody Affinity Purification	His-tag Protein Affinity Purification
Ligand or Chemistry (Catalog #)	DEAE: 9079 (5 ml) Q: 9081 (5 ml) CM: 9080 (5 ml)	Protein A: 9076 (1 ml) Protein G: 9077 (1 ml)	Ni-IMAC: 9078 (5 ml)
Base Support	DEAE: Highly cross-linked 6% Sepharose Q: Highly cross-linked 6% Sepharose CM: Highly cross-linked 6% Sepharose	Protein A: Rigid, highly cross-linked Agarose Protein G: Highly cross-linked 4% Sepharose	Highly cross-linked 6% Sepharose
Formats	A drip column / column chromatography	A drip column / column chromatography	A drip column / column chromatography
Binding Capacity (mg/ml)	DEAE: Weak anion exchange 110-160 µmol Cl- /ml media Q: Strong anion exchange 180-250 µmol Cl- /ml media CM: Weak cation exchange 90-130 µmol H+/ml media	Protein A: 60 mg (human IgG)/ml media Protein G: ≥ 20 mg (human IgG/ml media	≥ 40 mg (His-tag protein)/ml media 16-23 µmol (Ni2+) /ml media
Equivalent Products	DEAE Sepharose 4FF Q Sepharose 4FF CM Sepharose 4FF	MabSelect sure LX Protein G Sepharose 4FF	Ni Sepharose 4FF

Affinity of Protein A/G for Immunoglobulin Types from Different Species

Protein A and Protein G exhibit specific binding affinities for various immunoglobulin (Ig) subtypes, subclasses, and species, which are determined by the characteristics of the Ig Fc region. The details of their binding affinities are listed in the following chart that categorizes the interaction strengths of Protein A and G with immunoglobulin subclasses across different species.

Species	Immunoglobulin	Binding to Protein A	Binding to Protein G
Human	IgG (normal)	++++	++++
	IgG1	++++	++++
	IgG2	++++	++++
	IgG3	-	++++
	IgG4	++++	++++
	IgM	-	-
	IgA	-	-
	IgE	-	-
Mouse	IgG1	+	++++
	IgG2a	++++	++++
	IgG2b	+++	+++
	IgG3	++	+++
Rat	IgG1	-	+
	IgG2a	-	++++
	IgG2b	-	++
	IgG2c	+	++
Goat	IgG	+/-	++
Rabbit	IgG	++++	+++
Sheep	IgG	+/-	++

"-" : No binding, "+" : Weak binding, "++" : Binding, "+++" : Moderate Binding, "++++" : Strong Binding

References

1. P. M. Cummins, K. D. Rochfort, B. F. O'Connor, Ion-Exchange Chromatography: Basic Principles and Application. *Methods Mol. Biol*. **1485**, 209–223 (2017).
2. A. Jungbauer, R. Hahn, "Chapter 22 Ion-Exchange Chromatography" in Methods in Enzymology, R. R. Burgess, Deutscher, Murray P., Eds. (Academic Press, 2009) vol. 463, pp. 349–371.
3. M. Urh, D. Simpson, K. Zhao, Affinity chromatography: general methods. *Methods Enzymol*. **463**, 417–438 (2009).