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NuGel™ Poly-Carboxy

Polymer Coated Silica Affinity Matrices

Special Features of NuGel™:

- Non-specific sites are virtually eliminated by a polymer coating
- Stable across a wide pH range 2 - 10
- 1000Å, 50µm Silica suitable for LC and batch processes

Special Features of Poly-Carboxy ligand:

- Covalently couples ligands containing free amino groups in the presence of a carbodiimide.
- Covalently couples non-polar ligands in organic solvents.
- pH stable from 2 to 9.

Silica has been an industry standard as an advantageous matrix suitable for high performance liquid chromatography. With NuGel™, non-specific sites have been virtually eliminated making it an ideal support for affinity purification. Through a proprietary polymer coating, Silica is cross linked forming a reactive Poly-Epoxy functionality stable across a wide pH range (pH 2 to 10). From this foundational chemistry, all of the NuGel™ affinity products are derived.

For Immobilization of Proteins, Antibodies, Hormones, Peptides, Haptens, Drugs, Etc.						
Product Name	**Matrix Reactive Group**	**Ligand Reactive Group**	**Special Features**	**Size**	**Column Volume (Approx)**	**Item No.**
NuGel™ Poly-Epoxy	Terminal Epoxy	Amino	Direct Coupling of Amino Groups	25 Grams	50 ml	NPEY-25
NuGel™ Poly-Amine	Terminal Amine	Carboxylic Acid, or Carbohydrate	Carbodiiamide reaction, or $NaIO_4$ derived Aldehyde	25 Grams	50 ml	NPAM-25
NuGel™ Poly-Aldehyde	Terminal Aldehyde	Amino	Direct Coupling of Amino Groups	25 Grams	50 ml	NPAY-25
NuGel™ Poly-Hydroxy	Terminal Glycol	Amino	Carbodiimidazole mediated reaction	25 Grams	50 ml	NPHX-25
NuGel™ Poly-Carboxy	Terminal Carboxylic Acid	Amino	Carbodiiamide mediated reaction	25 Grams	50 ml	NPCY-25

* Kilogram quantities and other particle sizes and porosity of NuGel™ are also available upon request.

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NuGel™ Poly-Carboxy Protocol

NuGel™ Poly-Carboxy is a derivative of NuGel™ Poly-Epoxy affinity support. This affinity support contains carboxy groups at the end of hydrophilic spacer arms and is used to couple ligands containing amino groups.

Technical Data	
Spacer Arm	Polymerized hydrophilic carbon chain
Porosity	1000Å
Average Particle Size	50um
Substitution Level	100-200 uEq/gm of carboxy groups

Special Features:

- Couples ligands containing free amino groups in the presence of a carbodiimide.
- Couples non-polar ligands in organic solvents.
- pH stable from 2 to 9.

Carbodiimide-Mediated Protocol for Aqueous Coupling of Poly-Amine to Carboxy-Containing Ligands Or Poly-Carboxy to Amine-Containing Ligands

1. Carbodiiamides can be used to facilitate the formation of amide bonds between carboxylate groups and amines. For protein coupling, water soluble EDC {1-ethyl-3-(3-dimethylamino-proplyl)Carbodiimide} is used; solvent soluble Carbodiimide such as DCC are also available. For protein-ligand, optimal coupling takes place under high protein concentrations, 10-20 mg/ml, but good results can be achieved with 1-2 mg/ml. Typical protein coupling ranges from (10 to 20) mg per ml column volume. Most literature references give an optimum reaction pH of 4.75, but any pH between 4.5 and 7.5 should work well. A suitable coupling buffer is 0.1 M MES, pH 4.75.

2. One gram of NuGel™ produces approximately 2 ml column (or bed) volume. Weigh out required amount and wash on a sintered glass funnel with DI water containing 0.1 M NaCl. Transfer to mixing vessel.

3. Transfer the protein-ligand solution to the washed NuGel™. Add 60 mg EDC per Gram of NuGel™ into suspension. Mix by orbital shaker or overhead stirrer. Do not use magnetic stirrer. Mix at room temperature for 3 hours.

4. Using a filter or column, wash the coupled suspension with cold coupling buffer. Wash the gel extensively with aqueous 0.1 M NaCl. Store at 4°C in a well-sealed container.

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Operating Modes

Since the support matrix is based on a rigid 50 µm particle, NuGel™ can be operated in low pressure pump or gravity flow columns, or in batch mode.

Related NuGel™ References

Patents

Monoclonal antibodies directed to the cytotoxic lymphocyte maturation factor European Patent EP0790255

Purification of immunoglobulins using affinity chromatography and peptide US 2006/0153834 A1

Affinity

Chaumet, Alexandre, Sandrine Castella, Laïla Gasmi, Aurélie Fradin, Gilles Clodic, Gérard Bolbach, Robert Poulhe, Philippe Denoulet, and Jean-Christophe Larcher. "Proteomic analysis of Interleukin enhancer binding factor 3 (Ilf3) and Nuclear Factor 90 (NF90) interactome." *Biochimie* (2013).

Dermot Walls, Robert McGrath and Sinéad T.Loughran A Digest of Protein Purification. *Methods Molecular Biology.* Volume 681: 3-23 (2011)

Ehrlich, G. K., Michel, H., Chokshi, H. P. and Malick, A. W. Affinity purification and characterization of an anti-PEG IgM. *Journal of Molecular Recognition*, 22: 99–103 (2009).

Development of hepatitis B virus capsids into a whole-chain protein antigen display platform: New particulate Lyme disease vaccines. *International Journal of Medical Microbiology* Volume 298, Issues 1-2, 3 January 2008, Pages 135-142

A sensitive and high-throughput assay to detect low-abundance proteins in serum Hongtao Zhang, Xin Cheng, Mark Richter & Mark I Greene. *Nature Medicine* 12, 473 - 477 (2006)

Transformation of a L-peptide epitope into a D-peptide analog. *Peptides Frontiers of Peptide Science American Peptide Symposia*, 2002, Volume 5, Session XI, 769-770

Expression and folding of an antibody fragment selected in vivo for high expression levels in Escherichia coli cytoplasm. *Research in Microbiology* Volume 153, Issue 7, September 2002, Pages 469-474

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Identification of model peptides as affinity ligands for the purification of humanized monoclonal antibodies by means of phage display *Journal of Biochemical and Biophysical Methods* Volume 49, Issues 1-3.2001

George K. Ehrlich, Pascal Bailon, Wolfgang Berthold. Phage Display Technology - Identification of Peptides as Model Ligands for Affinity Chromatography *Affinity Chromatography Methods in Molecular Biology*, 2000, Volume 147, 209-220

A Digest of Protein Purification and partial amino acid sequence of a 28 kDa cyclophilin-like component of the rat liver sigma receptor. *Life Sciences* , Volume 55, Issue 8, 1994.

Nachman, M., Azad, A. R. M. and Bailon, P. (1992), Efficient recovery of recombinant proteins using membrane-based immunoaffinity chromatography (MIC). *Biotechnology and Bioengineering*, 40: 564–571.

Kinetic aspects of membrane-based immunoaffinity chromatography. Journal of Chromatography A Volume 597, Issues 1-2, 24 April 1992, Pages 167-172

Identification of model peptides as affinity ligands for the purification of humanized monoclonal antibodies by means of phage display. Methods in Molecular Biology, 2000, Volume 147, 209-220

Membrane-based receptor affinity chromatography. Journal of Chromatography A Volume 597, Issues 1-2, 24 April 1992, Pages 155-166 9th International Symposium on Affinity Chromatography and Biological Recognition

Ion Exchange

Levin W Protein Purification of recombinant human secretory phospholipase A2 (group II) produced in long-term immobilized cell culture. *Expr Purif* 1992 Feb;3(1):27-35.

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