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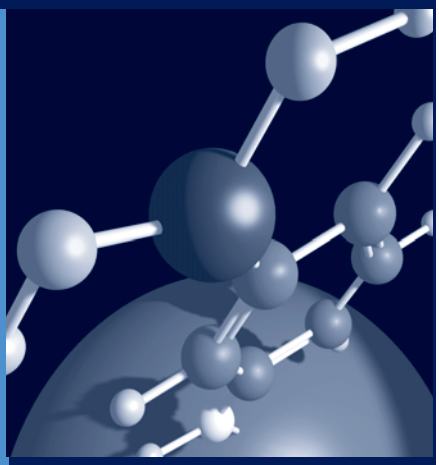

- *High recovery*
- *High throughput*
- *Wide process flexibility*

DATA SHEET

ProSep®-PB Chromatography Media

For the affinity chromatography of a variety of glycoproteins, nucleic acids, and other biomolecules

ProSep-PB media is designed for the affinity chromatography of a variety of glycoproteins, nucleic acids, and other biomolecules containing 1,2 diol functionality. ProSep-PB media can be used in the purification of glycoproteins, nucleotides and viruses.

ProSep-PB media is composed of a synthetic m-aminophenyl ligand immobilized on controlled pore size glass beads. The non-compressible nature of the media and the low back pressure generated, allows efficient media utilization at all scales.

Phenyl boronate ligand mediated chromatography differs in one fundamental way from most other ligands used for affinity chromatography. Whereas most ligands bind to a particular binding site on a protein by a mixture of non-covalent interactions, phenyl boronate interacts predominantly by forming a temporary covalent bond with 1,2-cis-diol groups. The boronate ligand will bind to any molecule containing the appropriate group, for example, erythropoiten (EPO), which is highly glycosylated. Steric configuration is only important in how it affects the accessibility of the 1,2-cis-diol groups. The generality of the binding mechanism makes ProSep-PB a versatile chromatography medium.

Examples of Affinity Chromatography Using Phenyl Boronate as a Ligand

TARGET MOLECULE	
Carbohydrates	Arabinose [7], fructose [1,7,20], galactose [4,20], glucose [1,4,7,20], inositol[1,20], lactose[4,7], mannitol[1,20], mannose[1,7,14], ribose[1,4,7], sorbitol[1,20], sucrose[1,20]
Nucleosides/Nucleotides	Adenosine [1,2,3,7,11,12,20], deoxyadenosine[1,7,16,20], 5′-AMP[2,6,7,20,22], 5′-ADP[2,6,7,20,22], 5′-ATP[2,6,7,20,22], cytidine[1,2,3,11,20], guanosine[1,3,11,20], thymidine[1,2,20], uridine[1,2,3,11,20], NAD(H)[15,22], NADP(H)[20,22]
Nucleic Acids	Oligonucleotides [2,5,18], tRNA aminoacylated from nonaminoacylated forms [5,8,18,19]
Peptides and Proteins	Erythropoiten (EPO)[23], membrane glycoproteins[21]
Other Small Molecules	Dopamine [10,13, 17], 3-methoxytyramine [13], norepinephrine [10,17], tyrosine [9,13]

Specifications	
Matrix	Porous glass with particle size in the range ≥ 74 ≤ 125 microns
Working pH Range	pH 2.5 – 8.5
Leachables	0.3ppm Si at pH7, decreasing at lower pH (0.01 ppm at pH 1.5). ProSep-PB media is stable between pH1 and pH9.
Pressure	Incompressible matrix with linear pressure/flow rate characteristics. Maximum operating pressure > 3000 psi (200 bar)
Shelf Life	1 year
Life	ProSep-PB media is stable over repeated operational cycles provided that proper cleaning protocols are observed.

References

1. Weith, H., Wiebers, j., and Gilham, P. 1970. Synthesis of cellulose derivatives containing the dihydroxyboryl group and a study of their capacity to form specific complexes with sugars and nucleic acid components. ***Biochem.*** 9:4396
2. Rosenberg, M., Weibers, J., and Gilham, P. 1972. Studies on the interactions of nucleotides, polynucleotides and nucleic acids with dihydroxyboryl-substituted celluloses. ***Biochem.*** 11:3623
3. Schott, H., 1972. New dihydroxyboryl-substituted polymers for column chromatographic separation of ribonucleoside-deoxyribonucleoside mixtures. ***Agnew. Chem.*** 84:819
4. Reske, K., and Schott, H. 1973. Column chromatographic separation of neutral sugars on a dihydroboryl-substituted polymer. ***Agnew. Chem.,*** 85:417
5. Schott, H., et al. 1973. A dihydroxyboryl-substituted methacrylic polymer for the column chromatographic separation of mononucleotides, oligonucleotides, and transfer ribonucleic acid. ***Biochem.***, 12:932
6. Moore, E., et al. 1974. Separation of ribonucleotides and deoxyribonucleotides on columns of borate covalently linked to cellulose. Application to the assay of ribonucleoside diphosphate reductase. ***Biochem.***, 13:2904
7. Yurkevich, A., et al. 1975. Study of the interaction of polyols with polymers containing N-substituted [(4-boronophenyl) methyl] – ammonio groups. ***Carbohyd. Res.***, 43:215
8. McCutchan, T., Gilham, P., and Soll, D. 1975. An improved method for the purification of RNA by chromatography on dihydroxyboryl substituted cellulose. ***Nucleic Acids Res.***, 2:853.
9. Ellinger, C., Chan, B., and Stanley, W. 1975. P-Vinylbenzeneboronic acid polymers for the separation of vicinal diols. ***J. Chrom.***, 104:57
10. Higa, S., et al. 1977. Isolation of catecholamines in biological fluids by boric acid gel. ***Anal. Biochem.***, 77:18
11. Uziel, M., Smith, L., and Taylor, S. 1976. Modified nucleosides in urine: selective removal and analysis. ***Clin. Chem.***, 22:1451
12. Davis, G., et al. 1977. HPLC separation and quantitation of nucleosides in urine and some other biological fluids. ***Clin. Chem.***, 23:1427
13. Hansson, C., et al. 1978. Chromatographic separation of catecholic amino acids and catecholamines on immobilised phenylboronic acid. ***J. Chrom.***, 161:352
14. Wulff, G., and Vesper, W. 1978. Preparation of chromatographic sorbents with chiral cavities for racemic resolution. ***J. Chrom.***, 167:171
15. Maestas, R., et al. 1980. Polyacrylamide-boronate beads saturated with biomolecules: A new general support for affinity chroma-tography of enzymes. ***J. Chrom.***, 189:225
16. Zimmerman, T., Wolberg, G., Duncan, G., and Elion, G. 1980. Adenosine analogues as substrates and inhibitors of S-adenosylhomocysteine hydrolase in intact lymphocytes. ***Biochem.***, 19:2252.
17. Gelijkens, C., and DeLeenheer, A. 1980. Simple method for isolating free urinary catecholamines by boric acid gel chromatography. ***J. Chrom.***, 183:78
18. Pace, B., and Pace, N. 1980. The chromatography of RNA and oligoribonucleotides on borate-substituted agarose and polyacrylamide. ***Anal. Biochem.***,107:128
19. Singhal, R., Bajaj, R., Buess, C., Smoll, D. and Vakharia, V. 1980. Reversed-phase boronate chromatography for the separation of o-methyl-ribose nucleosides and amino-t-RNA's. ***Anal. Biochem.***, 109:1
20. Glad, M., et al. 1980. High performance liquid affinity chromatography of nucleosides, nucleotides and carbohydrates with boronic acid substituted micrparticulate silica. ***J. Chrom.***, 200:254
21. Williams, G., Johnstone, A., Bouriotis, V., and Dean, P. 1981. Affinity chromatography of membrane proteins on dihydroxyboryl-matrix gel. ***Biochem. Soc. Trans.***, 9:137
22. Bouriotis, V., Galpin, I., and Dean, P. 1981. Applications of immobilised phenylboronic acids as supports for group specific ligands in the affinity chromatography of dehydrogenases. ***J. Chrom.***
23. D. Zanette, A. Soffientini, C. Sottani, E. Sarubbi. 2003. Evaluation of phenylboronate agarose for industrial scale purification of erythropoiten (EPO) from mammalian cultures. ***J. Biotech 101,*** 275-287

Ordering Information

Description	Pack Size	Former CAE Reference No.	Catalogue No.
ProSep-PB Media	10 mL	CAE 004124	113 247 324
	50 mL	CAE 004126	113 247 326
	100 mL	CAE 004127	113 247 327
	500 mL	CAE 004129	113 247 329
	1000 mL	CAE 004130	113 247 330
	5 L	CAE 004133	113 247 333
	10 L	CAE 004135	113 247 335

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