

AFFINITY PURIFICATION

Profinia™ Buffers for Protein A and Protein G Chromatography

The Profinia protein purification system offers preprogrammed chromatography methods for antibody purification using Protein A and Protein G cartridges. The Profinia Protein A and G methods allow simple, fast, and automated purification of antibodies and are fully compatible with cartridges available from Bio-Rad and a number of commercial vendors. Users can customize the methods and buffers according to their individual needs, but specific buffer recommendations are listed in Tables 1 and 2.

Table 1. Recommended buffers and solutions for Protein A chromatography using the Profinia Protein A and G plus desalting method.

Buffers and Solutions	Composition	Volume*	Port #	Catalog #
5x Protein A binding buffer** (Profinia desalting buffer)	685 mM NaCl, 13.5 mM KCl, 21.5 mM Na ₂ HPO ₄ , 40.5 mM KH ₂ HP0 ₄ , pH 7.0	200 ml	B1	620-0216
5x Protein A elution buffer (citrate)	500 mM sodium citrate, pH 3.0	—	B3	—
5x Protein A elution buffer*** (alternative: glycine)	500 mM glycine HCl, pH 2.5	—	B3	—
5x Profinia desalting buffer**	685 mM NaCl, 13.5 mM KCl, 21.5 mM Na ₂ HPO ₄ , 40.5 mM KH ₂ HP0 ₄ , pH 7.0	200 ml	B4	620-0216
2x Profinia cleaning solution 1**	1 M NaCl, 100 mM Tris, pH 8.0	125 ml	B5	620-0217
4x Profinia cleaning solution 2**	2 M NaCl, 400 mM CH ₃ COONa, pH 4.5	125 ml	B6	620-0218
2x Profinia storage solution** (desalting cartridge)	4% C ₆ H ₅ CH ₂ OH (benzyl alcohol)	200 ml	B7	620-0219
1x 20% ethanol (Protein A cartridge storage buffer)	20% ethanol (v/v)	—	B8	—

*Volumes reflect purchasable amounts. ** Also available as part of the Profinia desalting purification kit (catalog #620-0228).

*** 5x glycine buffer at 1,250 mM and pH 3.0 is also compatible with integrated desalting methods.

Note: The above solutions should be used at the concentrations shown. The Profinia protein purification system automatically dilutes buffers to 1x working solutions.

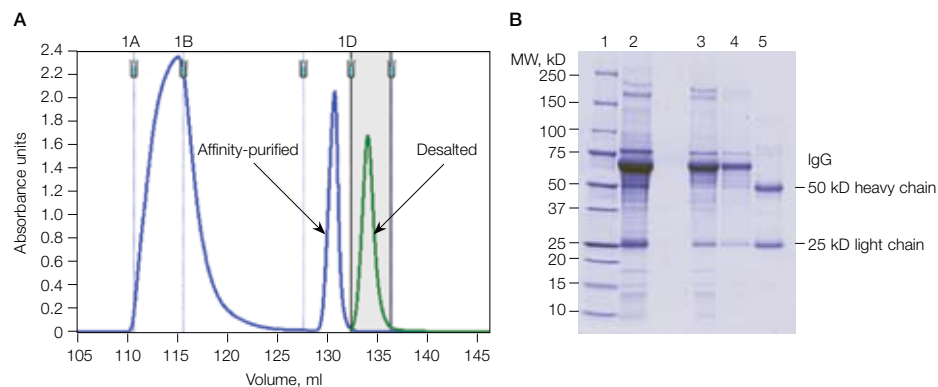


Fig 1. Purification of IgG fraction from 1 ml human serum using 1 ml Bio-Scale™ Mini Affi-Prep® Protein A and 10 ml Bio-Scale™ Mini Bio-Gel® P-6 desalting cartridges with the recommended buffers in table 1. A, chromatogram showing protein A affinity purification (–) and desalting (–) profiles; 1A, flowthrough; 1B, wash; 1D, affinity-purified and desalted IgG; **B**, SDS-PAGE of chromatography fractions. Lane 1, Precision Plus Protein™ standards; lane 2, load; lane 3, flowthrough (1A); lane 4, wash (1B); lane 5, elution (1D).



Table 2. Recommended buffers and solutions for Protein G chromatography.*

Buffers and Solutions	Composition	Volume**	Port #	Catalog #
5x Protein G binding buffer	100 mM sodium phosphate, pH 7.0	—	B1	—
5x Protein G elution buffer (glycine)	500 mM glycine HCl, pH 2.7	—	B3	—
5x Profinia desalting buffer***	685 mM NaCl, 13.5 mM KCl, 21.5 mM Na ₂ HPO ₄ , 40.5 mM KH ₂ HPO ₄ , pH 7.0	200 ml	B4	620-0216
2x Profinia cleaning solution 1***	1 M NaCl, 100 mM Tris, pH 8.0	125 ml	B5	620-0217
4x Profinia cleaning solution 2***	2 M NaCl, 400 mM CH ₃ COONa, pH 4.5	125 ml	B6	620-0218
2x Profinia storage solution*** (desalting cartridge)	4% C ₆ H ₅ CH ₂ OH (benzyl alcohol)	200 ml	B7	620-0219
1x 20% ethanol (Protein G cartridge storage buffer)	20% ethanol (v/v)	—	B8	—

* Protein G chromatography also uses the Profinia Protein A and G plus desalting method. ** Volumes reflect purchasable amounts. *** Also available as part of the Profinia desalting purification kit (catalog #620-0228).

Note: The above solutions should be used at concentrations shown. The Profinia protein purification system automatically dilutes buffers to 1x working solutions. The protein A and G methods are compatible with 1 ml and 5 ml cartridges available from a number of commercial vendors.

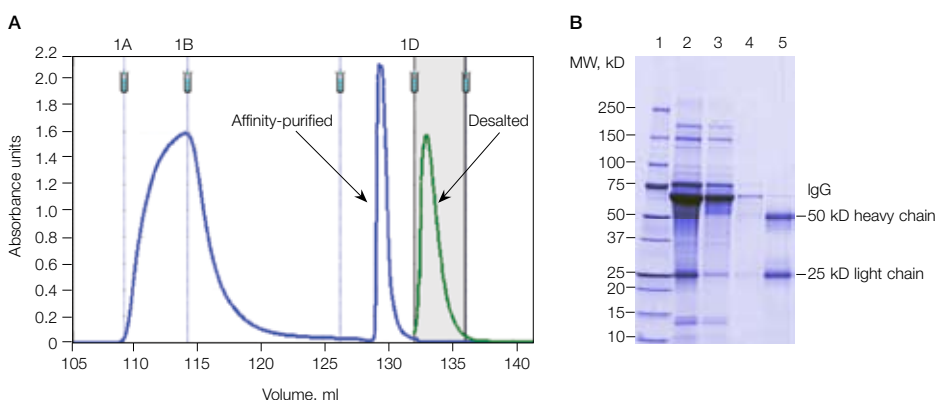


Fig 2. Purification of IgG fraction from 1 ml mouse serum using 1 ml HiTrap Protein G HP column and 10 ml Bio-Scale Mini Bio-Gel P-6 desalting cartridge with the recommended buffers in table 2. A, chromatogram showing different fractions; 1A, flowthrough; 1B, wash; 1D, affinity-purified and desalted IgG; B, SDS-PAGE of chromatography fractions. Lane 1, Precision Plus Protein standards; lane 2, load; lane 3, flowthrough (1A); lane 4, wash (1B); lane 5, elution (1D).

Ordering Information

Catalog # Description

Prepacked Bio-Scale Mini Cartridges

620-0216	5x Profinia Desalting Buffer, 200 ml
620-0217	2x Profinia Cleaning Solution 1, 125 ml
620-0218	4x Profinia Cleaning Solution 2, 125 ml
620-0219	2x Profinia Storage Solution, 200 ml
620-0224	Profinia Desalting Buffer Kit, includes purification buffers, cleaning and storage solutions; sufficient for 10 applications
620-0228	Profinia Desalting Purification Kit, 10 ml, includes Profinia desalting buffer kit, 2 x 10 ml desalting cartridges
732-4600	Bio-Scale Mini Affi-Prep Protein A Cartridges, 5 x 1 ml
732-4602	Bio-Scale Mini Affi-Prep Protein A Cartridges, 1 x 5 ml
732-5304	Bio-Scale Mini Bio-Gel P-6 Desalting Cartridges, 5 x 10 ml
732-5314	Bio-Scale Mini Bio-Gel P-6 Desalting Cartridges, 5 x 50 ml
732-5312	Bio-Scale Mini Bio-Gel P-6 Desalting Cartridge, 1 x 50 ml

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