

Protein A, IgG-Fc binding protein

Product Description

Name:	Protein A , from <i>Staphylococcus aureus</i> Purified	Recombinant (<i>E. coli</i>)
Catalog number:	275755 , 1mg	402909, 2mg 40290A, 10mg 402490B, 100 mg
Molecular Weight:	MW= 42 KDa (55-56KDa by SDS-PAGE)	MW= 45 KDa
Form:	Lyophilized	
Specific activity:	Protein A will bind the Fc portion of IgGs from many species including most human and mouse subclasses, and Rabbit, Cat, Pig, Monkey, Horse Igs. It does not recognize Human IgG3, IgE and weakly IgA and IgM, Mouse IgM and IgG1 (see table FT-408992).	
Applications:	Indirect coating for ELISA and other immunoassays. Unlabeled protein A is also used to prepare ou labelled Ab-probes for immunoassays, and affinity supports for affinity purification of antibodies, and immunoprecipitation.	
Storage:	Store at +4°C (-20°C possible for long term storage (K))	

Scientific and Technical Information

Protein A is a highly stable surface receptor produced by *Staphylococcus aureus*, of 42 kD in its native form which is capable of binding the Fc portion of immunoglobulins, especially IgGs, from a large number of laboratory or domestic species (Boyle, 1987).

- Protein A can bind up to 5 molecules of IgG, allowing the formation of a precipitate (Sjöholm, 1975) . Optimal binding occurs at pH 8.2, with high-affinity ($K_a = 10^8$ /mole). Isoelectric point is 4.85-5.10. It is stable in pH 1.0-12.0 range. Extinction coefficient is $E_{280}(1\%) = 1.4$.

- Recombinant Protein A is highly purified (produced in *E. coli*), a monomeric protein lacking cysteine residue. >95% pure by SDS-PAGE analyses; A_{280} of 0.1% solution: -0.165

Endotoxin activity: less than 0.1 EU/μg of Protein A as determined by LAL method.

- Protein A will bind the Fc portion of human IgG subclasses and mouse IgG_{2a} and IgG_{2b}, and IgG₁ (weak). Protein A also binds IgGs Rabbit, Sheep, Bovine, Goat, Monkey IgGs, Rat IgGs (but not IgG_{2b}), and Horse IgG. It does not recognize or weakly Human and Mouse IgM, IgA, IgE or serum albumin.

- As Protein A does not bind bovine IgGs, it is taken to good account for the purification of monoclonal antibodies from FCS supplemented culture media (where [Protein G #75194A](#) co-binds unspecific bovine IgGs). However, several monoclonal antibodies do not bind to Protein A, especially the majority of rat immunoglobulins and mouse IgG1 ([Kronvall 1970](#)).

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FT-40290A

See more protein A/G/L information properties in sheet FT-[40290A](#).

Applications

Antibody binding proteins such as Protein A and Protein G are often used in solid-phase immunoassays (coating), and to prepare affinity supports for antibody purification and related techniques such as immunoprecipitation (IP) and chromatin immunoprecipitation (ChIP).

Below are some guidelines for use.

In any case, it is important to match the binding specificity of the immobilized antibody binding protein to the species and antibody subtype of the antibody that you wish to capture: each antibody binding protein varies in its ability to bind to antibodies of different subtypes and species.

See the technical sheet [FT-408992](#) for the comparison table of Protein A/G/L affinity toward Ig subclasses and species. See also other Ig-binding protein, such as jacalin.

Reagents preparation

After reconstitution, Protein A may be stored at +4°C (stable several days) or -20°C for long term, in working aliquots; no appreciable decay in Fc-binding activity has been detected after six months of storage.

Coating Protein A/G/L to polystyrene microplates

Non labeled products are used typically for coating purpose in solid-phase immunoassays.

1- Dissolve the protein L at 1mg/ml in distilled water or PBS.

Note: The solubility can be improved by addition of glycerol up to 10%, without affecting its binding properties.

2- Prepare a 10 µg/ml solution in 0.1 M carbonate buffer pH 9.6

3- Pipette 100µl of Protein L solution into microplates wells and incubate overnight at +4°C (with a cover or an adhesive film on the plate)

4- Wash wells with PBS (Phosphate 100mM, NaCl 150mM, pH 7.5)

5- Pipette 200µl per well of saturating agent (1% BSA (v/v) in PBS or TBS - see related products) and incubate for 2 Hrs at 37°C.

6- Wash twice with PBS (Phosphate 100mM, NaCl 150mM, pH 7.5)
Plates can be used immediately or in following days.

Antibody purification

See the technical sheets of Agarose-Protein A.

Literature – Protein A

Boyle, M. D. P. and K. J. Reis. Bacterial Fc Receptors. *Biotechnology* 5:697-703 (1987).

Sjöholm, I. Protein A from *Staphylococcus aureus*. Spectropolarometric and spectrophotometric studies. *Eur. J. Biochem.* 51:55-61 (1975).

Watson, P. R., Guthrie, T. H. and Caruana, R. J. Cisplatin-associated hemolytic-uremic syndrome--Successful treatment with a *Staphylococcal* Protein A column. *Cancer* 64:1400-1403 (1989).

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FT-40290A

Related products and documents

- Other Ig Binding proteins and immunoreagents:

Protein A, G, L immunoreagent	Protein A conj.	Protein G conj.	Protein L conj.
> Unlabeled	Protein A #40290A	Protein G #75194A.	Protein L #56874A
> Labeled proteins A, G, L:			
HRP (peroxidase)	408992,	751952,	L79611
AP (Alkaline Phosphatase)	804102	80413A	
Biotin (Avidin Binding Protein)	303970	43791A	BI0300
FITC [492/520nm]	408974	43853A	
SR101 [596/620nm]	692512		
TRITC [550/570nm]	685652	438671,	
R-PE (PhycoErythrin) [488/580nm]	CE8010	CE8030	
APC (AlloPhycoCyanin) [633nm/670nm]		CE8020	
> Immobilized supports			
Agarose	UP49981	UP75196	52746G.

[Elution buffers](#) (for IgG, #Q99542)

JACALIN (Artocarpus Integrifolia Lectin – from Jackfruit) [#52280A](#) and its conjugates

- **Saturating agents:**

Carbonate coating Buffer [#R16490](#)

BSA agents: 30% solution [#UP900101](#), powders [#Q84171](#), BSA-containing [PBS&TBS](#) buffers

SeaBlock agent [#UP40301A](#) (to reduce background due mammalian crossreactivities)

- See [BioSciences Innovations catalogue](#) and [e-search tool](#).

Other information

For use *in vitro* research use only, not for diagnostic. Not for use in diagnostic or therapeutic procedures.

For any information, please contact your local distributor.

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rev : S1E-M11E-H05E

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