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G-Biosciences ♦ 1-800-628-7730 ♦ 1-314-991-6034 ♦ technical@GBiosciences.com

A Geno Technology, Inc. (USA) brand name

Well-Coated™ Protein L

96-Well Plates Coated with
Immunoglobulin Binding Protein L

(Cat. # 786-737, 786-776, 786-777)



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INTRODUCTION

Well-Coated™ Protein L plates are designed to bind the kappa light chains of immunoglobulins without interfering with the antigen binding site. The Well-Coated™ Protein L plates offer greater range of immunoglobulin classes and subclasses compared to Protein A, Protein G and Protein A/G. Protein L will bind to all classes of IgG, including IgG, IgM, IgA, IgE and IgD, and binds to single chain variable fragments (scFv and Fab fragments).

Well-Coated™ Protein L plates are for single antibody assays and are not suitable for multiple assays (sandwich ELISAs) as the first antibody will not block all IgG binding sites and therefore false positives will occur with the second antibody. The wells are coated to a 100µl depth and are supplied pre-blocked in our proprietary Superior™ Blocking Buffer. The clear, white and black plates are offered for colorimetric, chemiluminescence and fluorescent detection systems, respectively.

KIT COMPONENTS

| Cat. # | Components | Size |
|---------|---|----------|
| 786-737 | Well-Coated™ Protein L Coated 8-well strip plate, Clear | 5 plates |
| 786-776 | Well-Coated™ Protein L Coated 96 well plate, Black | 5 plates |
| 786-777 | Well-Coated™ Protein L Coated 96 well plate, White | 5 plates |

STORAGE CONDITIONS

Shipped at ambient temperature. Upon arrival, store unopened at 4°C. Once opened the plates can be stored in a resealable bag (ZipLoc) with an appropriate desiccant at 4°C.

IMPORTANT PROTEIN L INFORMATION

- Only binds to immunoglobulins with type kappa I, III and IV in human and kappa I in mouse (see appendix)
- May be specific for certain kappa subgroups in other species
- Binds scFv without interfering with antigen binding
- Has weak binding affinity for rabbit immunoglobulins
- No binding affinity for bovine, goat or sheep immunoglobulins
- No binding affinity for lambda light chains

ADDITIONAL ITEMS REQUIRED

- Antibody to be bound to plate (see Appendix for correct *Well-Coated*[™] plate to be used).
- Wash Buffer: femtoTBST[™] (Cat. # 786-161) or femtoPBST[™] (Cat. # 786-162); 10X concentrated wash buffers supplemented with Tween[®] 20. Or an appropriate wash buffer of choice.
- Blocking Buffer: A suitable blocking buffer, we recommend our *Superior*[™] Blocking Buffer (Cat. # 786-655 to 786-661) or NAP-BLOCKER[™], an animal free blocking agent suitable for ELISA (Cat. # 786-190).
- Labeled Antigen, visit www.GBiosciences.com for horseradish peroxidase (HRP), alkaline phosphatase (AP) and biotin labeling kits.
- Detection system, femtoELISA[™] is a chromogenic detection system for HRP and AP (Cat. # 786-110 to 786-113)

PROTOCOL

The following protocol is a simple ELISA protocol and the protocol and reagents used will have to be optimized for specific applications and assays.

Basic ELISA Assay

1. Wash the wells to be used three times with 300µl Wash Buffer.
2. Dilute the antibody to be bound to ~1µg/ml with the Blocking Buffer. Add up to 100µl to each well.
3. Incubate at room temperature for 30-60 minutes, for optimal binding use a plate shaker.
4. Wash each well three times with 300µl Wash Buffer.
5. Add the labeled antigen at a concentration of ~0.1µg/well, diluted in Blocking Buffer, if necessary.
6. Incubate at 37°C for 1 hour.
7. Wash each well three times with 300µl Wash Buffer.
8. Detect the label signal according to the manufacturer's instructions, using 200µl detection reagent per well.

NOTE: For biotin, incubate the plate for a further 1 hour at 37°C with an enzyme-labeled streptavidin or other biotin detection system. Wash as before and then detect the signal.

APPENDIX

| Species | Antibody Class | Protein L | Species | Antibody Class | Protein L |
|----------------|-------------------|-----------|-------------------|------------------|-----------|
| Mouse | Total IgG | ***** | Human | Total IgG | ***** |
| | IgM | ***** | | IgG ₁ | ***** |
| | IgG ₁ | ***** | | IgG ₂ | ***** |
| | IgG _{2a} | ***** | | IgG ₃ | ***** |
| | IgG _{2b} | ***** | | IgG ₄ | ***** |
| | IgG ₃ | ***** | | IgM | ***** |
| Rat | Total IgG | ***** | | IgD | ***** |
| | IgG ₁ | ***** | | IgA | ***** |
| | IgG _{2a} | ***** | | Fab | ***** |
| | IgG _{2b} | ***** | | ScFv | ***** |
| | IgG _{2c} | ***** | Goat | Total IgG | - |
| Cat | Total IgG | ? | | IgG ₁ | - |
| Chicken | Total IgY | - | | IgG ₂ | - |
| Cow | Total IgG | - | Dog | Total IgG | ? |
| | IgG ₁ | - | Guinea Pig | Total IgG | ? |
| | IgG ₂ | - | Pig | Total IgG | ***** |
| Horse | Total IgG | ? | Rabbit | Total IgG | ** |
| | IgG(ab) | ? | Sheep | Total IgG | - |

Table 1: Relative affinity of Protein L for Immunoglobulins. The strength of binding only refers to immunoglobulins with the appropriate kappa light chains.

RELATED PRODUCTS

Download our Assay Development Handbook.



<http://info.gbiosciences.com/complete-assay-development-handbook>

For other related products, visit our website at www.GBiosciences.com or contact us.

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