Well-Coated™ Neutravidin™

96-Well Plates Coated with Neutravidin™
for Binding Biotinylated Molecules

(Cat. # 786-743, 786-766, 786-767)
INTRODUCTION
Well-Coated™ Neutravidin™ plates are designed to specifically bind biotinylated molecules, including biotin tagged antibodies, with minimal non-specific binding. This is particular advantageous for antibodies known to denature upon direct binding to polystyrene plates.

Biotin exhibits an extraordinary binding affinity for avidin ($K_a=10^{15} \text{M}^{-1}$) and Neutravidin™ ($K_a=10^{15} \text{M}^{-1}$). Biotin and avidin interaction is rapid and once the bond is established it can survive up to 3M guanidine-hydrochloride and extremes of pH. Biotin-avidin bonds can only be reversed by denaturing the avidin protein molecule with 8M guanidine-hydrochloride at pH1.5 or by autoclaving. Neutravidin™ is in many respects is similar to avidin except that it has no carbohydrate side chains to eliminate lectin binding; is of near neutral pl (6.3) to reduce non-specific adsorption; lacks the RYD sequence eliminating interaction with RGD domain of adhesion receptors. The binding of Neutravidin™ is similar to that of avidin and streptavidin with less non-specific binding.

Well-Coated™ Neutravidin™ plates are suitable for direct, indirect, competitive and sandwich assays. The wells are coated to a 200µl depth and are supplied pre-blocked in our proprietary Superior™ Blocking Buffer. The plates are protected with our WellCoat™ Stabilizer (Cat. # 786-1217) that creates a protective layer over the immobilized agents. The reagent will not interfere with the assay and has no effect of the efficiency or capacity of the wells. The WellCoat™ Stabilizer offers greater protection and shelf life of the plates. In some cases, the protective layer may give the appearance of a white coating. The clear, white and black plates are offered for colorimetric, chemiluminescence and fluorescent detection systems, respectively.

ITEMS) Supplied

<table>
<thead>
<tr>
<th>Cat. #</th>
<th>Components</th>
<th>Size</th>
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<tbody>
<tr>
<td>786-743</td>
<td>Well-Coated™ Neutravidin™ Coated 8-well strip plate, Clear</td>
<td>5 plates</td>
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<tr>
<td>786-766</td>
<td>Well-Coated™ Neutravidin™ Coated 96 well plate, Black</td>
<td>5 plates</td>
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<tr>
<td>786-767</td>
<td>Well-Coated™ Neutravidin™ Coated 96 well plate, White</td>
<td>5 plates</td>
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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.

BINDING CAPACITY
*Well-Coated™ Neutravidin™*: ~40pmol D-biotin/well
ADDITIONAL ITEMS REQUIRED

• Biotinylated antibody (10µg/ml) to be bound to plate; visit www.GBiosciences.com for biotin labeling kits.
• 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).
• Antigen
• Enzyme Labeled Primary Antibody; visit www.GBiosciences.com for horseradish peroxidase (HRP) and alkaline phosphatase (AP) 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 direct ELISA protocol and the protocol and reagents used will have to be optimized for specific applications and assays.

Direct ELISA Assay
1. Wash the wells to be used two times with 300µl Wash Buffer.
2. Add up to 200µl biotinylated sample to each well.
3. Incubate at room temperature for 1-2 hours, for optimal binding use a plate shaker.
4. Wash each well three times with 300µl Wash Buffer.
5. Make serial dilutions of the antigen, diluted in Blocking Buffer, and add 200µl to each well.
6. Incubate at room temperature for 0.5-1 hour with shaking.
7. Wash each well three times with 300µl Wash Buffer.
8. Add 200µl enzyme labeled primary antibody.
9. Incubate at room temperature for 0.5-1 hour with shaking.
10. Wash each well five times with 300µl Wash Buffer.
11. Detect the label signal according to the manufacturer’s instructions, using 200µl detection reagent per well.

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.