INSTRUCTIONS

Pierce® Streptavidin High Binding Capacity Coated 96-Well Plates

15500 15501 15502 15503 0920.2

Number	Description
15500	**Pierce Streptavidin High Binding Capacity Coated Plates (clear, 96-well),** 5 each
15501	**Pierce Streptavidin High Binding Capacity Coated Plates (clear, 8-well strips),** 5 each
15502	**Pierce Streptavidin High Binding Capacity Coated Plates (white, 96-well),** 5 each
15503	**Pierce Streptavidin High Binding Capacity Coated Plates (black, 96-well),** 5 each
	Blocking Buffer: Plates are supplied blocked with SuperBlock® Blocking Buffer
	Binding Capacity: ~125pmol D-biotin/well
	Coating Volume: 100µL
	Blocking Volume: 200µL

Storage: Upon receipt store plates at 4°C in unopened pouches. Once opened, place unused plates in a resealable bag with desiccant and store at 4°C.

Introduction

The Thermo Scientific Pierce Streptavidin High Binding Capacity Coated Plates are made of polystyrene and ideal for binding assays using biotinylated molecules. These plates are especially advantageous when direct adsorption to polystyrene plates denatures antibodies or the target molecule. Streptavidin has no carbohydrate groups and an isoelectric point of 5-6, resulting in low nonspecific interactions. The Streptavidin Coated Plates are available in clear for colorimetric assays, white for chemiluminescent assays, and black for fluorescent assays.

Our proprietary coating technology is used to create high binding capacity (HBC) Pierce Streptavidin Coated Plates. These HBC plates provide a wider detection range and better curve linearity for small biotinylated ligands, such as peptides and oligonucleotides, than the standard coated plates.

Example ELISA Procedure

The following protocol describes a generalized enzyme-linked immunosorbent assay using a biotinylated capture antibody. Please see the reference list for other possible applications using streptavidin-coated microplates.

A. Materials Required

- Wash Buffer: Tris-buffered saline (25mM Tris, 150mM NaCl; pH 7.2; Product No. 28376), 0.1% BSA, 0.05% Tween®-20 Detergent; alternatively, use Thermo Scientific Blocker BSA (Product No. 37520) supplemented with 0.05% Tween-20 Detergent
- Biotinylated capture antibody adjusted to 10µg/mL, or other appropriate concentration, with Wash Buffer
- Antigen adjusted to appropriate concentration with Wash Buffer
- Primary antibody adjusted to appropriate concentration with Wash Buffer
- Enzyme-labeled secondary antibody adjusted to appropriate concentration with Wash Buffer
- Appropriate enzyme substrate: example substrates are the Thermo Scientific TMB Substrate Kit (Product No. 34021) for horseradish peroxidase and the Phosphatase Substrate Kit (Product No. 37620) for alkaline phosphatase

B. Method

1. Wash each well three times with 200µL of Wash Buffer. Add 100µL of the biotinylated capture antibody to each well and incubate for 2 hours with shaking at room temperature.
2. Wash each well three times with 200µL of Wash Buffer. Make a serial dilution of the antigen and add 100µL to each well. Incubate plate for 30 minutes with shaking at room temperature.
3. Wash each well three times with 200µL of Wash Buffer. Add 100µL of the primary antibody to each well and incubate plate for 30 minutes at room temperature.
4. Wash each well three times with 200µL of Wash Buffer. Add 100µL of the enzyme-labeled secondary antibody to each well. Incubate plate for 30 minutes with shaking at room temperature.
5. Wash each well three times with 200µL of Wash Buffer.
6. Follow the manufacturer's instructions for the specific detection system.

Related Thermo Scientific Products

37070	**SuperSignal® ELISA Pico Chemiluminescent Substrate,** 100mL
15159	**QuantaRed® Enhanced Chemifluorescent HRP Substrate Kit**
15169	**QuantaBlu™ Fluorogenic Peroxidase Substrate Kit**
34028	**1-Step™ Ultra TMB-ELISA,** 250mL
37621	**1-Step PNPP,** 100mL
29339	**Biotinylated Alkaline Phosphatase,** 1mg
29139	**Biotinylated Horseradish Peroxidase,** 5mg
15036	**Sealing Tape for 96-Well Plates,** 100/pkg
15520	**High Sensitivity Streptavidin Coated Plates (clear),** 5/pkg
15530	**High Sensitivity NeutrAvidin™ Coated Plates (clear),** 5/pkg
15511	**Pierce NeutrAvidin® High Binding Capacity Coated Plates (clear, 384-well),** 5/pkg
15512	**Pierce NeutrAvidin High Binding Capacity Coated Plates (white, 384-well),** 5/pkg
15513	**Pierce NeutrAvidin High Binding Capacity Coated Plates (black, 384-well),** 5/pkg

General References

Denlinger, L.C., *et al.* (2001). Cutting Edge: The nucleotide receptor P2X7 contains multiple protein- and lipid-interaction motifs including a potential binding site for bacterial lipopolysaccharide. *J Immunol* **167:**1871-6.

Ferre-Aubineau, V., *et al.* (1995). Colorimetric microtiter plate hybridization assay using monoclonal antibody for detection of an amplified human immunodeficiency virus target. *J Virol Meth* **55:**145-51.

Hiller, Y. *et al.* (1987). Biotin binding to avidin. Oligosaccharide side chain not required for ligand association. *Biochem J* **248:**167-71.

Holmstrom, K., *et al.* (1993). A highly sensitive and fast non-radioactive method for detection of polymerase chain reaction products. *Anal Biochem* **209:**278-283.

Simon, M.D., *et al.* (2004). A phage display selection of engrailed homeodomain mutants and the importance of residue Q50. *Nucl Acid Res* **32(12):**3623-31.