Thermo Scientific Nunc Immobilizer Streptavidin
Instruction protocol

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The Thermo Scientific™ Nunc™ Immobilizer™ Streptavidin Plates/Strips are manufactured using a patented photochemical method[1] for covalent coupling of ligands to polymer surfaces. Streptavidin, a high affinity biotin-binding protein isolated from Streptomycetes avidinii, is covalently coupled via a spacer to the plastic plates/strips. Streptavidin has a molecular weight of approx. 60,000 Dalton and has an isoelectric point (pI) close to 5. The Nunc Immobilizer Streptavidin Plates/Strips are designed and optimized for detection of various types of biotinylated biomolecules like biotinylated oligo-nucleotides, peptides and proteins.

Materials:

- Nunc Immobilizer Streptavidin Plates/Strips
- Biotinylated molecule of choice

Reagents:

- 5 x SSCT, pH 7.0 (5 x SSC (750 mM NaCl, and 75 mM Sodium Citrate) containing 0.05% (v/v) TWEEN® 20)
- 2 x SSCT, pH 7.0 (2 x SSC (300 mM NaCl, and 30 mM Sodium Citrate) containing 0.05% (v/v) TWEEN 20)
- PBST, pH 7.2 (Phosphate Buffered Saline containing 0.05% (v/v) TWEEN 20)

Note: Buffers should be used within one week from preparation.

Recommended coupling concentrations:

- Biotinylated oligonucleotides: We recommend using the following concentration range: 0.01 – 0.5 µM diluted in 5 x SSCT buffer pH 7.0
- Biotinylated peptides: We recommend using the following concentration range: 1 ng/ml – 1 µg/ml diluted in a PBST buffer pH 7.2
- Biotinylated proteins: We recommend using the follow-ing concentration range: 0.05 µg/ml – 5 µg/ml diluted in a PBST buffer pH 7.2

Coupling protocol for 96 well plate, and 8 well strips:

1. Pre-wash your plate with 3 x 300 µl/well PBST or 5 x SSCT buffer without any incubation step. This is done to ensure improved readouts and a very low coefficient of variation (CV%)
2. Prepare a solution of your biotinylated molecule in PBST buffer or 5 x SSCT (oligonucleotides)
3. Add the solution to the wells of a Nunc Immobilizer Streptavidin plate/strip (100 µl/well)
4. Incubate the plate with gentle agitation for 1 hour at room temperature (20° - 25°C)
5. Aspirate the wells and wash with PBST or 2 x SSCT (oligonucleotides) 3 x 300 µl/well
6. Your surface is now ready for assay application

Coupling protocol for 384 well plate:

1. Pre-wash your plate with 3 x 100 µl/well PBST or 5 x SSCT buffer without any incubation step. This is done to ensure improved readouts and a very low coefficient of variation (CV%)
2. Prepare a solution of your biotinylated molecule in PBST buffer or 5 x SSCT buffer (oligonucleotides)
3. Add the solution to the wells of a Nunc Immobilizer Streptavidin plate (50 µl/well)
4. Incubate the plate with gentle agitation for one hour at room temperature (20° - 25°C)
5. Aspirate the wells and wash with PBST or 2 x SSCT (oligonucleotides) 3 x 100 µl/well
6. Your surface is now ready for assay application

Specifications:

- Streptavidin coated area ~100 µl/well (96 well format)
- Stable when stored at room temperature to the expiration date, which appears on the case label

Trademarks and patents:

TWEEN 20 is a registered trademark of ICI American Inc., U.S.A. Immobilizer is a trademark of Exiqon A/S, Vedbaek, Denmark.
The product is produced under license from Exiqon A/S - EP 08 20483 and foreign applications and patents

References:

[1] Koch, T., Jacobsen, N., Fensholdt, J., Boas, U., Fenger, M. Jakobsen, M. H., Photochemical Immobilization of Anthraquinone Conjugated Oligonucleotides and PCR Amplicons on Solid Surfaces. Bioconjugate Chem. 11 (2000), 474-483.

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