PerkinElmer Life and Analytical Sciences
710 Bridgeport Avenue
Shelton, CT 06484-4794 USA

Technical Data Sheet

For Laboratory Use. A research product for research purposes only.

Streptavidin FlashPlate® PLUS
96-well Streptavidin Coated FlashPlate Microplates
SMP103, SMP103A

Product Description

This product is a 96-well FlashPlate coated with Streptavidin. The interior of each well is permanently coated with a thin layer of polystyrene-based scintillant followed by covalent binding of streptavidin molecules and a blocking step to block non-specific binding sites. This provides a platform for high-throughput, non-separation assays using a variety of isotopes.

Product Application

Streptavidin FlashPlate PLUS microplates are suitable for a wide variety of assay applications using biotinylated capture molecules including enzyme assays (e.g. polymerase, kinase), protein binding assays and immunoassays. The plates can be used with a number of isotopes such as ^{125}I, ^{3}H, ^{14}C, ^{35}S and ^{33}P.

Materials Included

Streptavidin FlashPlate PLUS, 5 Plate Pack
Catalog #: SMP103
96-well microplates - 5
TopSeal™ Microplate Covers - 10
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Streptavidin FlashPlate PLUS, 20 Plate Pack
Catalog #: SMP103A
96-well microplates - 20
TopSeal Microplate Covers - 40
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Storage and Stability

Store plates at 2-8°C with desiccant and protect from direct sunlight. Plate is stable through the expiration date.

Equipment Required

FlashPlate microplates are designed for use with the PerkinElmer TopCount® Microplate Scintillation and Luminescence Counter. Software is pre-set for the various isotopes that may be used with FlashPlates. Refer to the instrument manual for detailed instructions on the appropriate settings to use.

The PerkinElmer MicroBeta® Trilux may also be used to count FlashPlate microplates. The bottom set of photomultiplier tubes must be shut off and the system programmed for the appropriate plate holder cassette.

Detailed information on the use of both instruments is available. Be sure that counting instrument is set for the appropriate isotope and counting conditions.

Coating Volume

The Streptavidin FlashPlate PLUS has been coated with a volume of 200 µL/well.

Performance Characteristics

Intra-Plate Precision

Within plate precision determined by assaying an IC_{50} concentration of [^{3}H]-biotin on entire plates from several different lots. Results are reported below:

Precision Analysis	Mean cpm	SD (cpm)	Pooled % CV	Pooled % CV + 2SD*
Data Set: n=58 plates	9090	564	5.4%	7.2%

*95% confidence interval

Inter-Plate Precision

Between plate precision was tested by assaying an IC_{50} concentration of [^{3}H]-biotin on plates from two different plate lots. Results of eight different experiments are reported below.

Experiment	Number of Plates Tested	Mean cpm	Inter-Plate Precision (% CV)
1	8	8097	3.3%
2	8	8696	3.8%
3	5	8816	3.3%
4	4	7893	4.4%
5	7	8927	2.3%
6	20	9473	3.5%
7	10	9164	2.1%
8	10	9340	3.3%

Binding Capacity

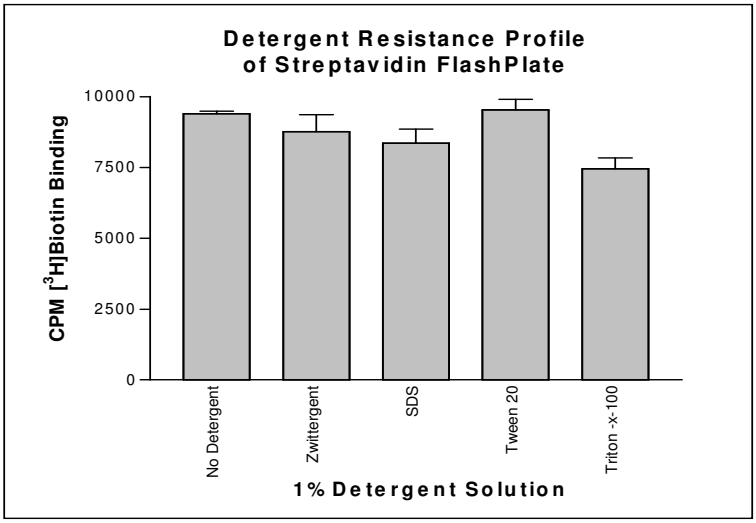
Binding capacity, as measured by a [^{3}H]-biotin binding assay, is typically ≥ 35 pmol per well. Actual binding capacity in an assay is dependent on the size of the biotinylated molecule.

Reagent Tolerances

The detergent and DMSO studies discussed below, demonstrated that the presence of these reagents, at certain concentrations, did not hinder biotin capture. However, these reagents may still interfere with specific assay systems. Each researcher must determine if his/her assay system is sensitive to these reagents.

Detergents

Several detergents were found to not interfere with the ability of Streptavidin FlashPlate to capture [^{3}H]-biotin. Detergent resistance was tested by assaying a solution of [^{3}H]-biotin at an IC_{50} concentration in PBS buffer alone, or PBS buffer with a final concentration of 1% detergent. Plates were incubated overnight at room temperature. The mean cpm values for each solution are plotted below.



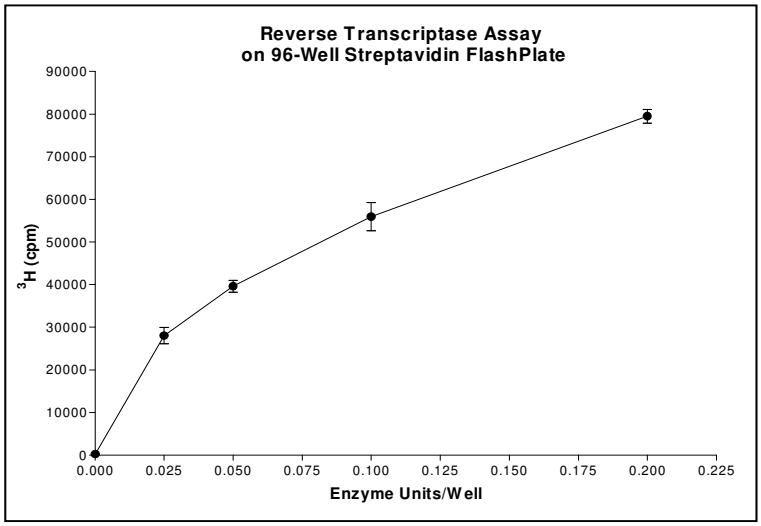
DMSO

DMSO concentrations up to 10% did not adversely affect the ability of Streptavidin FlashPlates to capture [^{3}H]-biotin. The direct addition of 100% DMSO to a Streptavidin FlashPlate will cause a significant loss in signal. This signal loss correlates with the volume of 100% DMSO added.

Application Examples

Reverse Transcriptase Assay

A reverse transcriptase assay was performed on a 96-well Streptavidin FlashPlate using recombinant HIV-1 reverse transcriptase (PerkinElmer Cat.# NEI490) and [methyl,1',2'-3H]dTTP (PerkinElmer Cat.# NET520A). Reaction solution containing tracer and dNTPs was added, followed by diluted enzyme. Plates were incubated overnight at 37°C and counted on a microplate scintillation counter. The background levels were less than 300 cpm in this assay.



Kinase Assay

The kinetics of the PKA phosphorylation of biotinylated kemptide (LRRASLG) was demonstrated on a Streptavidin FlashPlate. PKA was added to the plate at 5 and 10 units per well. The reaction was initiated by the addition of reaction buffer, substrate (500 pmol per well) and [γ-^{33}P]-ATP (PerkinElmer Cat.# NEG302H). The assay was incubated for the times indicated in the graph and the reaction terminated by an aspiration step followed by two washes with PBS. Plates were counted on a microplate scintillation counter.