



life line lab S.r.l

Streptavidin coated microplate



Protocol information

For Laboratory use only
Store desiccated at 2-8°C
Consistent results are obtained by precisely following the instructions below.



life line lab S.r.l

Via Castagnetta, 7
00040 Pomezia (RM)

Tel.: +39 06 91489209
Fax: +39 06 91489270
E-mail: info@lifelinelab.com

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5. Stability

Compliance with the IVDD requires that at least one batch of each product is subjected to stability testing after under-going the process of shipment.

It was therefore decided to perform a stability test on each batch of Streptavidin coated microplates after submitting them to 3 cycles of 12 hours exposure at 37°C, with a minimum period of 12 hours at 2°C to 8°C between each cycle.

Method:

After the initial treatment with 150 µl / well of physiological solution (see instruction before use), stressed and non stressed Streptavidin coated wells are incubated with biotinilated alkaline phosphatase (AP-biotin).

Excess unbound AP-biotin is removed by washing.

A substrate solution for AP (pNPP) is then added to each well.

Optical density read at 405 nm is used to determine stability.

Specification : Loss after simulated transport ≤ 15%

Availability

Streptavidin coated microplate

12 x 8 well strips

Product N° MP0100

3. *Binding capacity*

Streptavidin coated wells together with BSA saturated wells were incubated with known quantities of biotin.

At the end of this first reaction unbound biotin was measured in a competitive assay for the quantitative determination of biotin.

The difference between added and recovered amount is considered the binding capacity of coated Streptavidin well.

Specification : ≥ 6 pmoles

4. *Specificity*

In order to ensure specificity of reaction towards biotin, different concentrations of biotinylated and non biotinylated AP were incubated into Streptavidin coated wells and the results were compared.

Method:

After the initial treatment with 150 μ l / well of physiological solution (see instruction before use), coated Streptavidin is incubated with biotinylated alkaline phosphatase (AP-biotin) and non biotinylated AP

Excess unbound is removed by washing.

A substrate solution for AP (pNPP) is then added to each well.

Optical density read at 405 nm is used to determine specificity.

Specification : $\frac{\text{OD AP-biotin (150 ng/ml)}}{\text{OD AP (150 ng/ml)}} \geq 500$

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Introduction

Streptavidin is a tetrameric protein (M.W. 70.000) and each sub-unit can bind one molecule of biotin. The result is the strongest non covalent interaction between bio-molecules: 260 ± 20 pN (G.U Lee, Langmuir vol. 10, pp. 354-357, 1994).

Streptavidin coated microplates are designed to specifically bind biotinylated molecules (e.g. proteins, peptides polysaccharides, oligonucleotides, DNA fragments) in a great variety of applications where passive adsorption does not guarantee the required sensitivity, specificity and stability.

Instructions before use

- a) Allow microplate to reach room temperature before cutting aluminium bag.
- b) Remove but not discard the aluminium bag.
- c) Fill the required number of wells with at least $150 \mu\text{l}$ / well of physiological solution (0,9% NaCl w/v in distilled water).
- d) Leave microplate for at least 5 minutes, on the bench, at room temperature.
- e) Blot dry by inverting the microplate and tapping firmly onto absorbent paper. All residual physiological solution should be blotted dry.

Storage instructions

- a) Unopened microplate must be stored at $2 \div 8^\circ\text{C}$.
- b) Opened, unused microplate strips must be stored with the desiccant provided at $2 \div 8^\circ\text{C}$ in the original aluminium bag sealed with tape.

QC Analysis

1. *Within Batch reproducibility*

In order to ensure homogeneity of dispensing and washing during manufacturing process, one strip every fifteen plates, is sampled and tested for reactivity and precision.

Method:

After the initial treatment with $150 \mu\text{l}$ / well of physiological solution (see instruction before use), coated Streptavidin is incubated with biotinylated alkaline phosphatase (AP-biotin)

Excess unbound AP-biotin is removed by washing.

A substrate solution for AP (pNPP) is then added to each well.

Optical density read at 405 nm is used to determine CV%

Specification : $\text{CV}\% \leq 7$

2. *Within Plate reproducibility*

In order to ensure homogeneity of treatment between all the wells of one plate, one microplate is randomly sampled and tested for reactivity and precision.

Method:

After the initial treatment with $150 \mu\text{l}$ / well of physiological solution (see instruction before use), coated Streptavidin is incubated with biotinylated alkaline phosphatase (AP-biotin)

Excess unbound AP-biotin is removed by washing.

A substrate solution for AP (pNPP) is then added to each well.

Optical density read at 405 nm is used to determine CV%

Specification : $\text{CV}\% \leq 5$