

Streptavidin Coating Kit

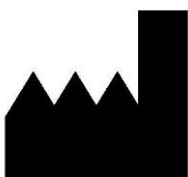
Instructions

Reagents for Streptavidin Coating incl. Blocking

Coating solution volume: 125 ml

For preparing coatings with a Biotin binding capacity of
the type "Standard Capacity"

Product Code: BTCK-SCB0125



BioTeZ Berlin-Buch GmbH
Robert-Rössle-Str. 10
13125 Berlin, Germany
Tel.: +49 (0) 30 9489 2130
Fax: +49 (0) 30 949 4509
E-mail: info@biotez.de
www.biotez.de

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Note: Read the instructions carefully before using the reagents!

Introduction

Streptavidin binds very tightly to the small molecule Biotin. Streptavidin coating of solid phases offers a universal immobilization principle for the detection and analysis of proteins, peptides, PCR-fragments, haptens etc., which must be present in biotinylated form.

As a further product BioTeZ offers a polymerized Streptavidin, the Polystreptavidin R, characterized by an extraordinary high Biotin binding capacity (MC coating = maximum capacity). Coatings made of Polystrept R combine the excellent binding capacity with a high chemo and thermo stability. It is very suitable for coating membranes, beads, biochips, plastics etc.





Streptavidin is part of this set of reagents, which has been supplemented by a special solution for pre-coating. The additional pre-coating serves to improve the outcome and should be used especially for very inert surfaces to ensure a high quality coating. The coating procedure starts with the pre-coating step.

Intended use

The product contains the reagents Streptavidin, Streptavidin Solution Buffer, Dilution Buffer Concentrate 10x and Blocking Solution.

It is used for adsorptive coating microplates, beads, membranes etc. made of nitrocellulose, ceramic or plastics like Polystyrene, Polyethylene, Polypropylene with Streptavidin "Standard Capacity".

Kit components 125ml Coating Solution

No.	Components*	Marking	Content / Volume	Color of top cap	Storage temperature
1	Streptavidin, lyophilized 5 mg*	SAV	2 x 5 mg	Colourless 	Upon arrival -20°C
2	Streptavidin Solution Buffer	SSB	1 x 3 ml	yellow 	+2 to +8°C
3	Dilution Buffer Concentrate 10x*	DBUF10x	1 x 14 ml	colourless 	+2 to +8°C
4	Blocking Solution	BS	1 x 125 ml	red 	+2 to +8°C

* Condition: dilution required

Scheme of coating procedure

For example coating of microplates with 100µl coating volume per well

Coating Step

Coating with **Streptavidin Coating Solution** in reconstituted **Dilution Buffer, 100µL/well**
Incubation: overnight (18 hours)
at room temperature (22 – 25°C)



Attention: No aspiration or washing steps are required after coating step.

Blocking Step

Blocking with **Blocking Solution 100µL/well**
Incubation: 15 minutes at room temperature (22 – 25°C,
(CV% values might be improved by gently stirring.)



Aspirating Step



Drying Step

3 hours or overnight at room temperature (maximum of 30°C) without covering of the microplate



Packing Step

Packing the microtiter plates in foil bags with desiccant and store at +2 to +8°C

Steps A. to G. for coating of microplates

A. Preparation of the reagents for coating

All reagents for the next step should be brought to room temperature (20-25 °C) before use. Then the Dilution Buffer Concentrate 10x has to be diluted with distilled water 1:10 (= reconstituted Dilution Buffer).

Volume of rec. Dilution Buffer	Volume of Dilution Buffer Concentrate 10x	Volume of distilled water
10	1 ml	9 ml
50	5 ml	45 ml
125	12.5 ml	112.5 ml

B. Solution of Streptavidin

The kit contains 2 vials with lyophilized recombinant Streptavidin. Each vial contains 5 mg Streptavidin.

Please prepare the solution of **Streptavidin concentrate**:

Add 1 ml **Streptavidin Solution Buffer** to the vial **Streptavidin** and allow the contents to dissolve for 5-10 minutes. Gently mix, but avoid foaming! After the solution the Streptavidin concentration is 5 mg/ml in this vial.

Please store the **Streptavidin concentrate** (5 mg/ml) at -20°C, if you don't use the entire amount.

C. Dilution of Streptavidin Concentrate for Coating Step

Attention: Depending on material and surface the suitable concentration of Streptavidin is to be tested out. The following concentration ranges are recommended:
10 – 30 µg/mL for coating of microplates
10 – 100 µg/mL for coating of membranes, beads etc.

The **Streptavidin Concentrate (5 mg/ml)** must be diluted immediately before use with the reconstituted **Dilution Buffer**.

For instance 30µg/ml Streptavidin Coating Solution as follows:

Volume of Streptavidin Coating Solution (30µg Streptavidin/ml)	Volume of Streptavidin Concentrate (5 mg/ml)	Volume of reconstituted Dilution Buffer
50	0.3 mL	49.7 mL
125	0.75 mL	124.25 mL

Pipette the appropriate volume of **Streptavidin Concentrate** into the appropriate volume of reconstituted **Dilution Buffer** and mix it.

The **Streptavidin Coating Solution** is now ready for coating step.

D. Coating Step

Dispense the appropriate volume e.g. 100µL of **Streptavidin Coating Solution** per well. Cover the plate with adhesive film and incubate at 22 - 25 °C for 18 hours (overnight).

Attention: No aspiration or washing steps are required after coating step. Do not empty the wells.

E. Blocking Step

Dispense the appropriate volume e.g. 100µL of **Blocking Solution** per well on the top of the Streptavidin coating solution. Cover the plate with adhesive film and incubate at 22 - 25 °C for 15 minutes by gently stirring. (CV% values might be improved by gently stirring.)

F. Aspirating Step

Suck off all wells of microtiter plate.

G. Drying Step

Dry the microtiter plates at 22 to 25°C (maximum of 30°C) for 3 h or overnight.

H. Packing Step

Pack the microtiter plates in foil bags with desiccant and store at 2 to 8°C.

Reagents that have exceeded their expiry date should be disposed of correctly.

Important points and precautions

1. The manufacturer assumes no liability and indicates that the user is solely responsible for the consequences of any alterations made, for non-observance of instructions or for performing the coating procedure without paying due attention.
2. The equipment used must be maintained in accordance with the manufacturers' instructions and any applicable guidelines. Before equipment is used it should be checked for fault-free operation.
3. Excess reagents that have exceeded their expiry date/lifetime should be disposed of correctly. You should observe the regulations that apply to you.
4. The coating reaction may only be carried out by trained specialists. The production steps are validated for use at the indicated temperature. Deviations in the climatic conditions can negatively influence the results.
5. Ensure that the materials, equipment and reagents are clean, paying particular attention e.g. to vessels and pipette tips.
6. Observe general health and safety regulations.
7. Follow the instructions for this test very closely. Pipette carefully into the wells, as otherwise excessive deviations in the results may occur.
8. The reagents contain substances such as sodium azide or Thiomersal as preservatives. They are toxic. Avoid contact with eyes and skin! Wear protective gloves!
9. When disposing of the reagents, observe any potential harm they may cause to the environment. Observe the regulations that apply to you.
10. Observe safety regulations, e.g. do not eat, drink or smoke in the workplace; keep materials and reagents away from foods and feeding stuffs; wear protective clothing (lab coat, safety glasses and gloves).
11. Never carry out pipetting operations using the mouth; always use suitable equipment or devices.



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<http://www.biotez.de>