

For life science research only.
Not for use in diagnostic procedures.



Blocking Reagent For ELISA

 **Version: 18**

Content Version: February 2021

Cat. No. 11 112 589 001 27 g
for one liter blocking solution
Not available in US

Store product at +2 to +8°C.

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1. General Information

1.1. Contents

Vial / Bottle	Label	Function / Description	Content
1	Blocking Reagent for ELISA	Powder, containing Tris buffer, sodium chloride, and a protein mixture that was obtained by proteolytic degradation of purified gelatin.	1 bottle, 27 g

1.2. Storage and Stability

Storage Conditions (Product)

When stored at +2 to +8°C or at +15 to +25°C, the product is stable through the expiration date printed on the label.

Vial / Bottle	Label	Storage
1	Blocking Reagent for ELISA	Store at +2 to +8°C or at +15 to +25°C. ⚠ Store dry.

Reconstitution

Dissolve the contents of one bottle in 100 ml double-distilled water at +15 to +25°C while stirring for approximately 30 minutes to obtain a clear solution.

⚠ Store autoclaved stock solution for several days to a week either unopened at +15 to +25°C or at +2 to +8°C after opening. Alternatively, store in aliquots at -15 to -25°C for up to 6 months.

1.3. Additional Equipment and Reagent required

Blocking of microplates

- Protein solution (antibody, antigen, etc.) at a concentration of 1 to 10 µg/ml in 50 mM sodium carbonate buffer, pH 9.2 to 9.4
- Wash solution: 150 mM NaCl, 0.1% Tween 20* (v/v) in double-distilled water

1.4. Application

Use the Blocking Reagent for ELISA for coating microplates. Blocking reagent usually yields significantly lower background levels than buffered bovine serum albumin solutions. Nevertheless it has to be taken into account that the protein components are gelatin hydrolysates.

⚠ Avoided using this blocking reagent when assays for monoclonal antibodies to collagen are performed.

2. How to Use this Product

2.1. Before you Begin

Working Solution

Dilute one aliquot of stock solution in a ratio of 1:10 with double-distilled water to yield a working solution containing 1% protein (w/v) in 50 mM Tris-HCl, 150 mM NaCl, pH approximately 7.4.

2.2. Protocols

Blocking of microplates

i See section, **Additional Equipment and Reagent Required** for more information.

- 1 Coat the wells of the microplate with 50 µl Protein solution.

- 2 Shake at 500 rpm for 0.5 to 2 hours at +15 to +25°C.
i *The optimal conditions may vary in this step. In some cases, coating at pH 7 and/or incubation overnight may be preferable.*

- 3 Pour out the solutions from the wells by inverting the microplate and by tapping it on a clean, dry cloth, such as cellulose to remove any adhering solutions.

- 4 Wash with 200 µl Wash solution per well.
 - Allow to take effect for 15 seconds, pour out, and tap.
 - Repeat washing step twice.

- 5 For postcoating, pipette 200 µl Blocking Reagent, working solution into each well and incubate for 15 minutes at +15 to +25°C.

- 6 Pour out the solutions from the wells by inverting the microplate and by tapping it on a clean, dry cloth, such as cellulose to remove any adhering solutions.

- 7 Wash with 200 µl Wash solution per well.
 - Allow to take effect for 15 seconds, pour out, and tap.
 - Repeat washing step twice.

- 8 Additional procedures are dependent on the assay and detection principle.

3. Supplementary Information

3.1. Conventions

To make information consistent and easier to read, the following text conventions and symbols are used in this document to highlight important information:

Text convention and symbols

 *Information Note: Additional information about the current topic or procedure.*

 **Important Note: Information critical to the success of the current procedure or use of the product.**

① ② ③ etc. Stages in a process that usually occur in the order listed.

① ② ③ etc. Steps in a procedure that must be performed in the order listed.

* (Asterisk) The Asterisk denotes a product available from Roche Diagnostics.

3.2. Changes to previous version

Layout changes.

Editorial changes.

3.3. Ordering Information

Product	Pack Size	Cat. No.
Reagents, kits		
Tween 20	50 ml, 5 x 10 ml	11 332 465 001

3. Supplementary Information

3.4. Trademarks

All product names and trademarks are the property of their respective owners.

3.5. License Disclaimer

For patent license limitations for individual products please refer to:

List of biochemical reagent products.

3.6. Regulatory Disclaimer

For life science research only. Not for use in diagnostic procedures.

3.7. Safety Data Sheet

Please follow the instructions in the Safety Data Sheet (SDS).

3.8. Contact and Support

To ask questions, solve problems, suggest enhancements or report new applications, please visit our **Online Technical Support Site.**

To call, write, fax, or email us, visit **sigma-aldrich.com**, and select your home country. Country-specific contact information will be displayed.

