

Bradford protein concentration assay kit

Item number: PC0010

Specification: 100T (2500 microholes)

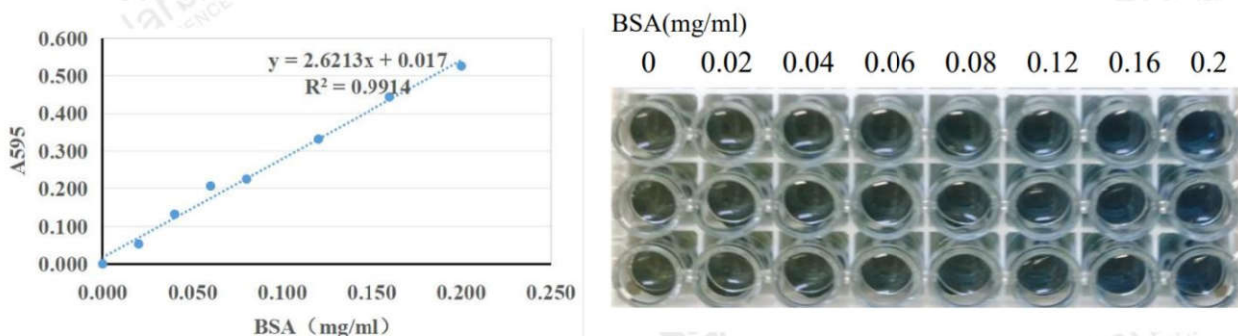
Storage: 2-8°C storage, valid for 12 months.

Product Contents:

Composition	Packaging (2500 microholes)	Save
5 x G250 stain solution	100mL	2-8 °C
PBS diluent	30mL	2-8 °C
Egg white standard (5mg/mL BSA)	1mL	-20 °C

Product description:

The Coomas Leilands G-250 dye, in an acidic solution, binds to the protein so that the position of the dye's maximum absorption peak (Imax), changes from 465nm to 595nm, within a certain concentration range, the determined absorbance value A595 is proportional to the protein concentration. Bradford method determination of protein concentration is not affected by most of the chemical substances in the sample, the concentration of merhydryl ethanol in the sample can be as high as 1M, the concentration of dithiothreitol can be as high as 5mM, but by a slightly high concentration of detergent, it is necessary to ensure that the concentration of SDS is less than 0.1%, Triton X-100 is less than 0.1%, And the Tween 20, 60 and 80 are below 0.06%. The BCA protein concentration assay kit produced by Solebao is recommended for samples containing detergents.



In Figure 1, the left figure shows the standard curve of the microplate method. The horizontal axis is the standard product of different concentration gradients, and the vertical axis is the absorption value at the corresponding 595nm. The figure on the right shows the actual color rendering effect of standard products with different concentration gradients. **Note: The data in the figure is for reference only, and the actual test results shall prevail.**

Operation instructions (for parameters only) :

I. Microporous enzyme labeling method

1. Completely dissolve the protein standard, take 10 μ L and dilute it to 250 μ L, so that the final concentration is 0.2mg/mL. In what solution the protein sample to be measured is in, the standard product should also be diluted with what solution. However, for the sake of simplicity, the standard can also be diluted with 0.9%NaCl or PBS.

Before use, please reverse and mix the 2.5 \times G250 dyeing solution 3-5 times. Take 1mL of the

2. 5 \times G250 dyeing solution, add 4mL of double steaming water, and mix well to form 1 \times G250

dyeing solution. The 1×G250 dyeing solution can be stored at 4°C for one week.

3 Add the standard product 0, 2, 4, 6, 8, 12, 16, 20 μl to the 96-well plate respectively, and add PBS diluent to make up to 20 μL, equivalent to the standard product concentration of 0, 0.02, 0.04, 0.06, 0.08, 0.12, 0.16, 0.2mg/mL

3. Dilute the sample appropriately (it is best to do several gradients, such as 2x, 4x, 8x dilution) and add 20 μL to the sample hole of the 96-well plate. Due to the error of the pipette when taking small amounts, the point in front of the standard line may not be very accurate, so as far as possible, let the sample point fall 1/2 behind the standard line.

5. Add 200 microliters of diluted 1 x G250 dye solution to each hole and let stand at room temperature for 3-5 minutes.

6. The absorbance of A595, or other wavelengths between 560-610nm, was measured with an enzyme label.

7. Calculate the protein concentration in the sample according to the standard curve.

II Spectrophotometer

if there is no enzyme label instrument, the dyeing reaction can be carried out in the centrifuge tube, the reaction liquid is mixed and added to the colorimetric dish, and the light absorption value is determined by spectrophotometer.

The steps are as follows:

1. Take eight (or more) clean 10mL centrifuge tubes and mark them with a number.

2. Take 100μL BSA and add it to PBS 2.4mL and dilute it until the final concentration is 0.2mg/mL.

3. Before use, please reverse the 3.5×G250 dyeing solution and mix it for 3-5 times. Take 10mL 5×G250 dyeing solution, add 40mL double steaming water, and mix it well to form 1×G250 dyeing solution. The 1×G250 dyeing solution can be stored at 4°C for one week.

4. Add the reagent according to the following table (measured in 5mL per well, the excess is used to clean the colorimetric dish), and the corresponding standard product concentration of the first 6 tubes is 0, 0.04, 0.08, 0.12, 0.16, 0.2mg/mL respectively

Centrifuge tube No.	1	2	3	4	5	6	7 (Sample tube 1)	8 (sample tube 2)	9 (sample tube 3)
Standard protein BSA	0μL	100μL	200μL	300μL	400μL	500μL	500μL appropriately diluted sample 1	500μL properly diluted sample 2
PBS	500 μL	400μL	300μL	200μL	100μL	0 μL	0μL	0μL	0μL
1XG250 dye solution	5mL	5mL	5mL	5mL	5mL	5mL	5mL	5mL	5mL

5 Measure the OD value after 3 minutes of reaction. For the accuracy of the experiment, a tube of dyeing solution can be added every 2 minutes, and a tube of OD value can be measured every 2 minutes. The following table:

Centrifugal tube number	1	2	3	4	5	6	7	8

Add dye solution (minutes)	0	2	4	6	8	10	12	14
Measure OD value	3	5	7	9	11	13	15	17

Related products:

- PC0001 BSA Standard (5mg/mL)
- PC0015 5×G250(for protein quantification)
- PC0021 BCA reagent
- PC0030 Lowry method protein concentration determination kit
- PC0020 BCA method protein concentration determination kit
- R0010 High efficiency RIPA tissue/cell fast lysate
- PR1600 prestain with low molecular weight protein MARKER
- R0050 nuclear protein extraction kit

Related literature:

- [1] Zhongyuan Li,Xiumei Li,Tianhui Liu,et al. The critical roles of exposed surface residues for the thermostability and halotolerance of a novel GH11 xylanase from the metagenomic library of a saline-alkaline soil. International Journal of Biological Macromolecules. July 2019; 133:316-323. (IF 4.784)
- [2] Qinlu Zhang,Qian Liu,Menghan Du,et al. Cetuximab and Doxorubicin loaded dextran-coated Fe₃O₄ magnetic nanoparticles as novel targeted nanocarriers for non-small cell lung cancer. Journal of Magnetism and Magnetic Materials. June 2018. (IF 3.046)

Note: Please refer to Solarbio website for more literature on the use of this product.