(FOR RESEARCH USE ONLY. DO NOT USE IT IN CLINICAL DIAGNOSIS!)

BCA Protein Assay Kit (BCA Method)

Catalog No.: BC00006

Size: 200T/600T

Please read the instructions carefully before use. If you have any questions or need further help during experiment, please don't hesitate to contact us through the following methods:

✉ Email (Sale) order@enkilife.com

✉ Email (Techsupport) techsupport@enkilife.com

☎ Tel: 0086-27-87002838

Website: www.enkilife.com

Shelf life: Please refer to the label on the outer package.

Techsupport: In order to provide you with better service, please inform us the lot number on the label of the outer package.

Basic Information

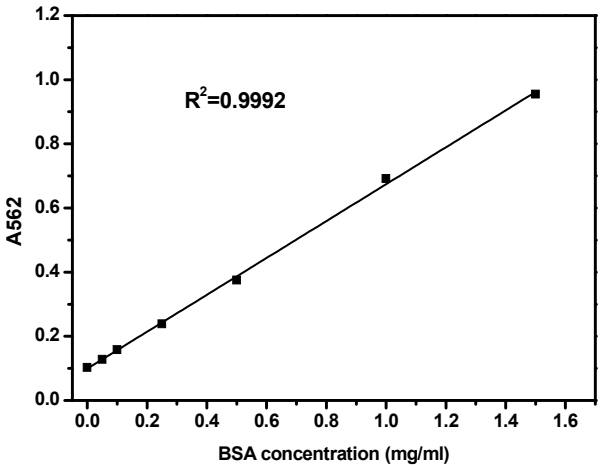
Product Name	BCA Protein Assay Kit (BCA Method)
Detection Method	Colorimetric
Sample Type	Serum,Plasma,Urine,Animal and Plant Tissues, Culture Medium,Cells
Assay Type	Quantitative
Detection Instrument	Microplate reader (540-595 nm, optimal detection wavelength 562 nm)

Product Introduction

This kit is a kit commonly used to determine the protein content in biological samples. The BCA method (Bicinchoninic Acid Method) is a method for determining protein concentration.

Principle

The BCA (Bicinchoninic Acid) protein content detection method is based on the ability of protein to reduce Cu^{2+} to Cu^{+} under alkaline conditions. Cu^{+} and BCA reagent can form a purple complex. The product has a characteristic absorption peak at 562 nm. The protein concentration of the sample can be quantitatively detected by the change in absorbance value.



Components

Serial number	Components	Size(200T)	Size(600T)	Storage
Reagent 1	BCA Reagent A	40ml	120ml	-20℃
Reagent 2	BCA Reagent B	1.2ml	1.2ml*3 vials	-20℃
Reagent 3	BSA Protein Standard	20 mg	20mg*3 vials	-20℃; after being prepared into solution, store at -20℃.
Reagent 4	Protein Standard Preparation Solution	1ml	1ml * 3 vials	-20℃
Consumable 1	Microplate(96 wells)	2 plates	6 plates	RT
Consumable 2	Plate Sealer	4 pieces	12 pieces	RT

Storage

The unopened kit can be stored at -20℃ for 12 months.

Experimential Preparation

1. Preparation of protein standards
Take 0.8 ml of protein standard solution and add it to a tube of protein standard (20 mg BSA). After fully dissolving, prepare a 25 mg/ml protein standard solution. Take an appropriate amount of 25 mg/ml protein standard and dilute it to a final concentration of 1 mg/ml. The dilution solution of the standard is generally the same as the solution of the sample to be tested. The standard can also be diluted with 0.9% NaCl or PBS. The protein standard solution and the diluted protein standard solution can be stored for a long time at -20ºC.
2. Preparation of BCA working solution
According to the number of samples, prepare an appropriate amount of BCA working solution by adding 50 volumes of BCA reagent A to 1 volume of BCA reagent B (50:1) and mix thoroughly. For example, add 5ml of BCA reagent A to 100μl of BCA reagent B, mix well, and prepare 5.1ml of BCA working solution. BCA working solution is stable at room temperature for 24 hours.

Operation process

1. 1 mg /ml standard solution was diluted in half to 0.5mg/ml , 0.25mg/ml , 0.125mg/ml , 0.0625mg/ml , and 0mg/ml (blank well), and 20 μl of each was added to the standard wells of a 96-well plate.
2. Add an appropriate volume of sample to the sample wells of the 96-well plate. If the

sample volume is less than 20μl, add standard diluent to make it up to 20μl.The protein concentration range determined by this kit is 0.02-1.5mg/ml.If the sample protein concentration is too high, dilution is required.

3. Add 200μl BCA working solution to each well and incubate at 37ºC for 30 minutes.
4. The absorbance at A562 or other wavelengths between 540-595 nm was measured using a microplate reader.

The operation table is as follows:

	Standard well	Assay well
Standards of different concentrations (μl)	20	--
Sample to be tested (μl)	--	20
BCA working solution (μl)	200	200
Incubate for 30 minutes at 37ºC with a microplate reader, and measure the OD value of each well at 562nm.		

Result calculation

Calculate the protein concentration of the sample based on the standard curve and the sample volume used.

Notes

1. A microplate reader is required, and the measurement wavelength is between 540-595nm , with 562nm being the best. Requires 96-well plate. If there is no microplate reader, you can also use an ordinary spectrophotometer for measurement, but when measuring, you need to appropriately increase the amount of BCA working solution according to the minimum detection volume of the cuvette so that it is not less than the minimum detection volume. The dosage can be scaled up accordingly or remain unchanged. When using a spectrophotometer to measure protein concentration, the number of samples that can be measured per kit may be significantly reduced.
2. The BCA method for determining protein concentration is not affected by the chemicals in most samples and is compatible with up to 5% SDS, 5% Triton X-100, and 5% Tween20, 60, and 80 in the sample. However, this kit is affected by chelator and slightly higher concentrations of reducing agents. It is necessary to ensure that EDTA is less than 10mM, there is no EGTA, dithiothreitol (DTT) is less than 1mM, and β-mercaptoethanol (β-Mercaptoethanol) is less than 0.01%. If the sample diluent or lysate itself has a high background, it will affect the accuracy of the kit.
3. When the BCA method is used to determine protein concentration, the color will deepen over time. The color development reaction will accelerate as the temperature rises. If the concentration is low, it is appropriate to incubate at a higher temperature or extend the

incubation time appropriately.
4. Unless the time and temperature of the color development reaction are precisely controlled, it is recommended to make a standard curve each time if you need to accurately determine the protein concentration.
5. This product is for Research Use Only, and shall not be used for clinical diagnosis or treatment, food or medicine, or stored in ordinary residences. .