BCA Protein Assay Kit

Cat#: 8013011/8013012

[Product Name]
BCA Protein Assay Kit

[Model & Specification]

Product Name	Cat No.	Size	Component No.	Component Name	Component Size	Quantity
BCA Protein Assay Kit	8013011	500 T	8013011-1	BCA Solution	100 mL	1 bottle
			8013011-2	Cu^{2+} Solution	2 mL	1 bottle
			8013011-3	Protein Standard （10 mg/mL）	1 mL	1 bottle
	8013012	5000 T	8013012-1	BCA Solution	200 mL	5 bottle
			8013012-2	Cu^{2+} Solution	4 mL	5 bottle
			8013012-3	Protein Standard （10 mg/mL）	1 mL	5 bottle

[Product Description]
BCA Protein Assay Kit is a sensitive and anti-interference total protein quantitative detection kit. Protein can reduce Cu^{2+} to Cu^{+} in an alkaline medium. BCA (Bicinchoninic acid) chelates with Cu^{+} to form a purple-blue chelate, and it can be determined by measuring the absorbance value of the chelate product at 562 nm to complete quantitative analysis of protein concentration. This kit is convenient and fast to use. After adding the sample to be tested, incubate for 30 minutes and measure the absorbance to complete the experiment. The linear range for the determination of protein concentration is 31.2 μg/mL~2000 μg/mL.
The kit can tolerate many detergent, including SDS, Triton X-100, Tween-20, etc.. As long as the concentration of the detergent is less than 5%, it will have no significant impact on the determination of protein concentration and is suitable for determination of protein concentration in cell and tissue protein samples extracted with RIPA lysis buffer.

[Storage And Transportation]
Protein standard is stored at -25℃~-15℃, and other components are stored at 2℃~8℃.
Transport on blue ice.

[Reagent Preparation]
1. Sample Preparation
(1) Cell Lysis
Collect the cells to be measured (1×10^6~1×10^7), add pre-cooled PBS to homogenate for lysis, or use medium-strength RIPA lysis buffer to lyse in an ice bath, centrifuge at 4℃ and 10,000 g for 10 minutes, and collect the supernatant for later use;
(2) Supernatant Solution of Tissue Lysis
Collect the tissue to be measured (20 mg~50 mg), add pre-cooled PBS or medium-strength RIPA lysis buffer, homogenize in an ice bath, centrifuge at 10,000 g for 10 minutes at 4℃, and collect the

supernatant for later use;
(3) Plasma
Take fresh anticoagulated blood and centrifuge it at 1,000 g for 10 minutes at 4℃. The supernatant solution is plasma;
(4) Serum
Take fresh blood, let it stand at room temperature for 30 minutes, and centrifuge at 2,000 g and 4℃ for 15 minutes. The supernatant solution is serum.
Note: If the sample can not measure immediately, please store them in a -80°C refrigerator. The kit involves the chelation reaction of Cu^{2+}, please avoid using chelating agents such as EDTA to treat the samples. If the chelating agent in the sample cannot to avoid, please keep the concentration ≤0.5 mM.
2. Preparation of Detection Working Solution
Prepare detection working solution by taking BCA Solution and Cu^{2+} Solution according to the proportions in the table.

	Volume Ratio （V：V）	1 sample	10 samples	100 samples
BCA Solution	50	200 μL	2 mL	20 mL
Cu^{2+} Solution	1	4 μL	40 μL	400 μL

3. Preparation of Protein Standard Solution
Dilute the Protein Standard (10 mg/mL) to 2000 μg/mL with the ddH_2O or the solution used to prepare the sample, and then dilute it twice into concentration gradients such as 1000, 500, 250, 125, 62.5, 31.2 μg/mL for standard curve determination.
It is recommended to prepare a new serie of dilutions of the standard before each measurement. If it cannot be used in time, it can be stored briefly at -25℃~-15℃ and used as soon as possible to avoid repeated freezing and thawing.
Standard Solution Preparation (Example):

No.	Concentration of standard solution (μg/mL)	Volume of standard solution (μL)	Volume of diluent(μL)	Total volume (μL)	Final Concen-tration (μg/mL)	Remaining volume(μL)
St8	10000	200	800	1000	2000	800
St7	2000	500	500	1000	1000	500
St6	1000	500	500	1000	500	500
St5	500	500	500	1000	250	500
St4	250	500	500	1000	125	500
St3	125	500	500	1000	62.5	500
St2	62.5	500	500	1000	31.25	1000
St1（Blank）	0	/	300	300	0	300

4. Measurement Method
(1) Refer to the table below and use a 96-well plate to conduct the experiment. Add protein standards (or samples) and detection working solution in sequence, then gently tap the 96-well plate or use a micropipette to mix the well plate with a constant temperature shaker. Place in an incubator and incubate at 37℃ for 30 minutes.

	Blank	Standard	Sample
The ddH_2O or the solution used to prepare the sample	20 μL	—	—

Protein Standard	—	20 μL	—
Sample	—	—	20 μL
Detection Working Solution	200 μL	200 μL	200 μL

(2) After the incubation, use a microplate reader to measure the absorbance at a wavelength of 562 nm.

Note: If the absorbance of the sample exceeds the absorbance value of the highest concentration of Protein Standard, the sample could be appropriately diluted before measurement. If the absorbance of the sample is too low, the sample could be appropriately concentrated before measurement.

5. Data Calculate

(1) Draw a standard curve for BCA protein concentration detection by preparing the concentration of the Protein Standard as the abscissa and the absorbance value as the ordinate.

(2) The BCA protein concentration detection standard curve is used to calculate the protein concentration in the sample.

(3) The protein concentration standard curve is as shown in the figure below:

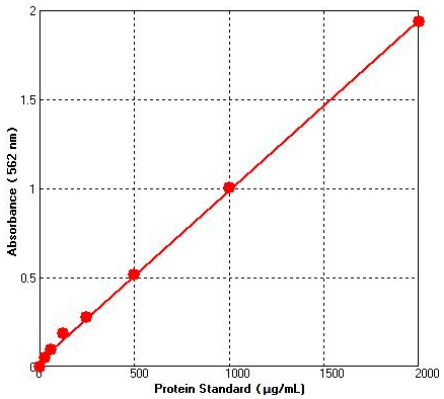

Figure 1. Protein Concentration Detection Standard Curve

Note: The standard curve is for reference only. Each experiment must prepare a standard curve for that experiment.

[Notice]

1. The kit involves redox reaction, and most oxidant and reducing agent will interfere with the determination of the kit. In particular, reagent containing thiol group such as DTT, mercaptoethanol and etc., which will seriously interfere with the measurement of this kit, so please avoid using them as much as possible.
2. The kit involves the chelation reaction of Cu^{2+}, please avoid using chelating agents such as EDTA to treat samples. If the presence of chelating agents in the sample cannot be avoided, keep the concentration ≤0.5 mM.
3. Each experiment must prepare a standard curve for that experiment.
4. When using the kit for the first time, please centrifuge the small volume liquid reagent appropriately before use.
5. The kit can only be used by professionals for scientific research, and may not be used for clinical diagnosis or treatment, and may not be used in food or medicine.
6. For your safety and health, please wear a lab coat and disposable gloves.

[Description Of Product Symbol]

Product Symbol	Description	Product Symbol	Description
REF	Catalog Number	LOT	Batch Code
	Date of Manufacture		Manufacturer

	Use-by date		Temperature limit
	Consult instructions for use		

[Company Information]
Manufacturer and after-sales service unit Name: Shenzhen Dakewe Bio-engineering Co., Ltd.
Website: www.dakewe.com
Telephone: (86-755) 86235300
Email: RD@dakewe.com
Address: Room 702-703, Building No.1, Shenzhen Biomedicine Innovations Industrial Park, No.14 Jinhui Road, Kengzi Street, Pingshan District, Shenzhen, China
After-sales service telephone: (86-755) 86235300
Zip Code: 518122