Product Manual

BCA Protein Assay Kit

Catalog No.: abx293001

Size: 100 tests / 500 tests / 2500 tests

Storage: Store the BSA solution at -20°C. Store Reagent A and Reagent B at 4°C for short-term storage, otherwise store at -20°C for long-term storage. Before use, warm the kit to room temperature.

Introduction

The BCA protein assay is a protein determination formulation based on bicinchoninic acid (BCA) for the colorimetric detection. This method combines the reduction of Cu2+ to Cu+ by proteins in an alkaline medium (the biuret reaction) and the soluble purple-colored reaction product from the complexing of Cu+ and BCA. This purple-colored complex exhibits a maximum absorbance at 562 nm that is nearly linear with increasing protein concentrations over a broad working range (0-2 mg/ml). Since it is highly sensitive and simple to use, the BCA protein assay is adopted by many laboratories and companies, and is one of the main protein quantitative methods as well as the Bradford assay.

Abbexa's BCA Protein Assay Kit has the characteristics of high sensitivity and light background, and the measured range can be up to 3 mg/ml.

Kit components (100 tests)

1. Reagent A: 20 ml
2. Reagent B: 1 ml
3. BSA (5 mg/ml): 0.4 ml

Kit components (500 tests)

1. Reagent A: 100 ml
2. Reagent B: 5 ml
3. BSA (5 mg/ml): 2 ml

Kit components (2500 tests)

1. Reagent A: 500 ml
2. Reagent B: 25 ml
3. BSA (5 mg/ml): 10 ml

Materials required but not provided

1. 37°C incubator
2. Spectrophotometer or plate reader (wavelength 562 nm)
3. Test tubes or 96-well plate
4. Distilled water (or reagent used to dilute samples)

A. Procedure

96 microplate procedure

1. Prepare the Working Reagent by mixing Reagent A and Reagent B (50:1) to produce a yellow-green solution.
2. Prepare the Standard Reagent by diluting the 5 mg/ml BSA standard into concentrations of 0.25, 0.5, 0.75, 1.0, 1.5 and 2.0 mg/ml. A control (0 mg/ml BSA) should also be prepared. The BSA dilutions can be frozen and stored at -20°C, and thawed and warmed to room temperature when used.
3. Mark the wells as Standard wells or Sample wells. Add 20 µl of the diluted Standard Reagent to each Standard well and 20 µl of sample to each Sample well.
4. Add 200 µl Working Reagent to each well and incubate for 30 min at 37°C.
5. Measure the absorbance at 562 nm on a plate reader. Subtract the average absorbance reading of at least 3 blank wells. Draw a standard curve by plotting absorbance reading for each BSA standard versus its concentration. Use the standard curve to determine the protein concentration of each unknown sample.

Test tube procedure

1. Prepare the Working Reagent by mixing Reagent A and Reagent B (50:1) to produce a yellow-green solution.
2. Prepare the Standard Reagent by diluting the 5 mg/ml BSA standard into concentrations of 0.25, 0.5, 0.75, 1.0, 1.5 and 2.0 mg/ml. A control (0 mg/ml BSA) should also be prepared. The BSA dilutions can be frozen and stored at -20°C, and thawed and warmed to room temperature when used.
3. Mark the tubes as Standard tubes or Sample tubes. Add 50 µl of the diluted Standard Reagent to each Standard tube and 50 µl of sample to each Sample tube.
4. Add 1 ml Working Reagent to each tube and incubate for 30 min at 37°C.
5. Measure the absorbance at 562 nm with a spectrophotometer. Subtract the average absorbance reading of at least 3 blank tubes. Draw a standard curve by plotting absorbance reading for each BSA standard versus its concentration. Use the standard curve to determine the protein concentration of each unknown sample.

Notes

1. Reagent A (BCA) should be a light yellow clear liquid. Discard Reagent A if discoloration or sediments are present.
2. Use Personal Protective Equipment (PPE) when carrying out the assay.
3. The BCA Protein Assay kit is not compatible with reducing or metal chelating reagents. If these reagents must be used, a Bradford Protein Assay kit is recommended.