

Protein Assay Kit (Colorimetric)

LS-K329-500 (500 Tests) • See Storage Conditions Below



Introduction

The protein is known as the "building blocks of life" and is one of the most important macromolecules in life science. Proteins are polypeptides made up of amino acids and play various key roles in all aspects of biology. Protein quantitation is a very common practice for life scientists. Simple, direct and automation-ready procedures for measuring protein concentration are very desirable. LSBio's protein assay kit is based on an improved Coomassie Blue G method. The dye forms a blue complex specifically with protein, and the intensity of color, measured at 595nm, is directly proportional to the protein concentration in the sample. The optimized formulation substantially reduces interference by substances in the raw samples and exhibits increased sensitivity towards peptides.

Key Features

- Sensitive and accurate. Use 10 mL samples. Detection range 0.06 - 1.0 mg /mL protein in 96-well plate assay.
- Simple and high-throughput. The "mix-and-read" procedure involves addition of a single working reagent and reading the optical density. Can be readily automated as a high-throughput assay in 96-well plates for thousands of samples per day.
- Low interference. Glucose, Tris, vitamins, and amino acids, DNA, RNA, salts, EDTA (< 12 mM), phenol (< 50 mM), urea (< 0.6 M), Triton (< 0.1%) and SDS (< 0.1% SDS) do not interfere in the assay.
- Versatility: assays can be executed in 96-well plate or cuvette.

Applications

- Direct Assays: total protein concentration.

Components

Component	K329-500
	500 Tests
Reagent (5 x concentrate)	20 mL
Protein standard (1.0 mg/mL BSA)	1 mL

Materials Not Supplied

Pipetting devices and accessories.

Procedure using 96-well plate: Blank 96-well plates (e.g. Corning Costar). Plate reader for 96-well plate.

Procedure using cuvette: Cuvettes and spectrophotometer.

Storage

The kit is shipped at room temperature. Store the reagent at 4°C and standard at -20°C, respectively. Shelf life: 12 months after receipt.

FOR RESEARCH USE ONLY! Not for use in humans.

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Assay Procedure

Reagent Preparation

Prepare enough working reagent by adding 1 vol of the 5 x Reagent to 5 vol of distilled water. Bring reagent to room temperature before use.

Procedure Using 96-Well Plate

1. Dilute standard as shown in the Table. Transfer 10 μ L diluted Standards and diluted sample in duplicate wells of a clear bottom 96-well plate. Store diluted standards at -20°C for future use.

No	Premix + H ₂ O	Vol (μ L)	BSA (mg/mL)	BSA (μ g/10 μ L)
1	100 μ L + 0 μ L	100	1.0	10
2	80 μ L + 20 μ L	100	0.8	8
3	60 μ L + 40 μ L	100	0.6	6
4	40 μ L + 60 μ L	100	0.4	4
5	30 μ L + 70 μ L	100	0.3	3
6	20 μ L + 80 μ L	100	0.2	2
7	10 μ L + 90 μ L	100	0.1	1
8	0 μ L + 100 μ L	100	0	0

2. Add 200 μ L working reagent and tap lightly to mix.
3. Measure OD at 570-630nm (peak 595nm).

Procedure Using Cuvette

1. Prepare standards as in the 96-well plate assay. Transfer 50 μ L diluted Standards and 50 μ L samples to cuvettes.
2. Add 1000 μ L working reagent and tap lightly to mix.
3. Measure OD at 570-630nm (peak 595nm).

Calculations

Subtract blank OD (water, #8) from the standard OD values and plot the OD against standard concentrations. Use the standard curve to determine the sample protein concentration.

General Considerations

If protein concentration is > 1 mg/mL, dilute samples in distilled water, and use OD values that lie within the calibration curve to calculate the sample protein concentration. Reading can be performed as soon as the reagent and sample are mixed. High sensitivity can be achieved by adding 50 μ L sample to 200 μ L Reagent (detection range 3 – 200 μ g/mL).

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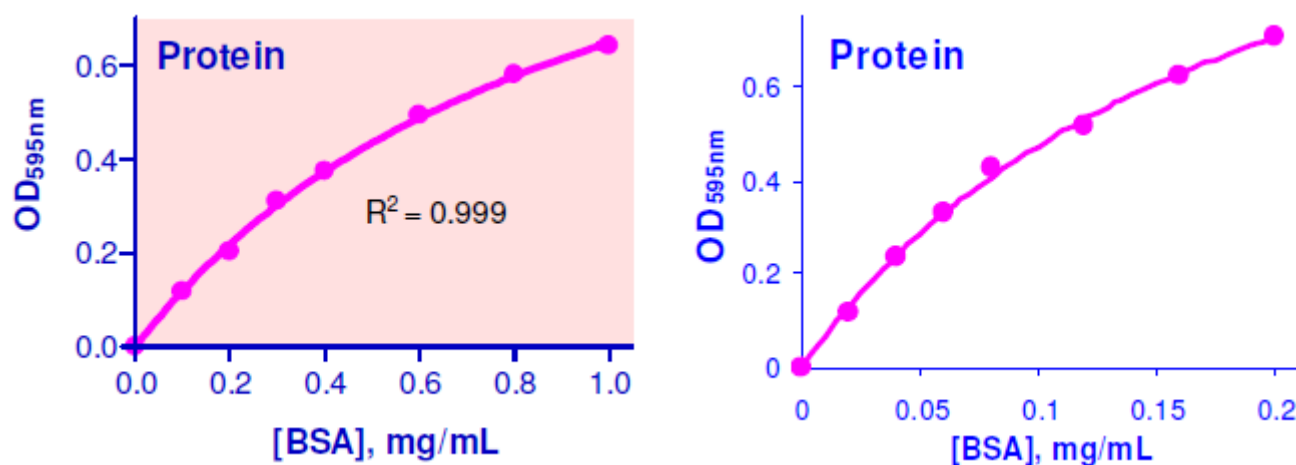
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Sample Data



Standard Curve in 96-well plate. Left: standard protocol. Right: 50 µL sample plus 200 µL Reagent.

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