

ab102535

Protein Quantitation Kit (Bradford Assay)

Instructions for Use

For the rapid, sensitive and accurate
measurement of Protein in various samples

This product is for research use only and is not
intended for diagnostic use.

Table of Contents

Table of Contents	2
1. Overview	3
2. Protocol Summary	3
3. Components and Storage	4
4. Assay Protocol	5
5. Data Analysis	8

1. Overview

Abcam's Protein Quantitation Kit (Bradford Assay) provides a simple and rapid procedure for determining the concentration of protein in solution. The method utilizes an improved Coomassie blue G reagent which forms a blue complex in the presence of protein. The intensity of the blue complex is proportional to the amount of protein in the sample and can be easily measured by spectrophotometer or plate reader at 595 nm.

2. Protocol Summary

Prepare 1X Working Solution



Dilute the Protein Standard I



Add Working Solution



Measure Optical Density

3. Components and Storage

A. Kit Components

Item	Quantity
5X Protein Assay Solution	20 mL
Protein Standard I	1.5 mL

* Store kit at +4°C or -20°C, protect from light. The BSA standard should be aliquoted after the first thaw and stored at -20°C. All reagents are stable for up to 12 months under proper storage conditions.

B. Additional Materials Required

- Microcentrifuge
- Pipettes and pipette tips
- Colorimetric microplate reader or spectrophotometer
- 96 well plate
- Orbital shaker

4. Assay Protocol

1. Prepare 1X working solution by diluting the 5X Protein Assay Solution with distilled water. Only prepare enough 1X working solution for a one-day experiment. Allow the solution to reach room temperature before use. The 1X working solution is stable for 24 hours. Left-over 1X solution should be discarded.
2. Depending on the volume and expected concentrations of your samples, either dilute the Protein Standard I (1 mg/mL) as in Table 1 or as in Table 2. Table 1 is appropriate for expected sample concentration ranges of 100 – 500 µg/mL and Table 2 is appropriate for expected sample concentration ranges of 2.5 – 25 µg/mL. If your expected sample concentration matches the concentration range in Table 2, predilute the Protein Standard I 1/10 with distilled water before following the dilutions in Table 2.
3. For Table 1, transfer 10ul diluted standards and 10ul diluted samples into duplicate wells in a clear bottom 96 well plate.

For Table 2, transfer 100ul diluted standards and 100ul diluted samples into duplicate wells in a clear bottom 96 well plate.

Table 1. Serial Dilutions for expected sample concentration range of 100 – 500 µg/mL and 10 µL sample volumes

No.	Standard (µl)	H ₂ O(µl)	Final Conc. (µg/mL)
1	0	50	0
2	5	45	100
3	10	40	200
4	15	35	300
5	20	30	400
6	25	25	500

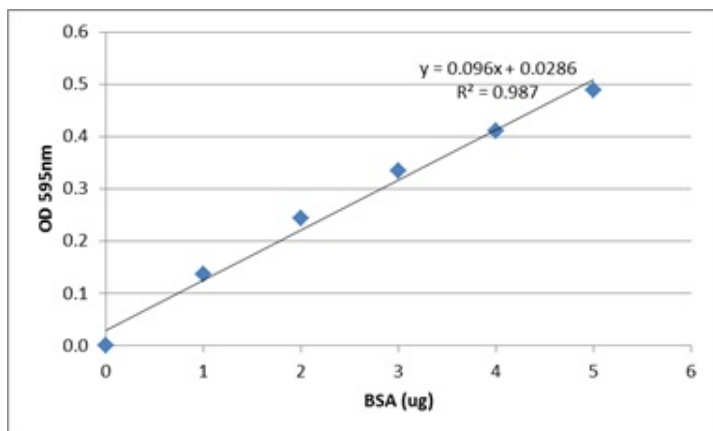
Table 2. Serial Dilutions for expected sample concentration range of 2.5 – 25 µg/mL and 100 µL sample volumes

No.	Standard (µL) (prediluted 1/10)	H ₂ O(µL)	Final Conc. (µg/mL)
1	0	1000	0
2	25	975	2.5
3	50	950	5
4	100	900	10
5	150	850	15
6	200	800	20
7	250	750	25

4. Add 100 μL of 1X working solution into each well that contains the standard and samples. Shake gently to mix. Incubate for 5 min at room temperature.
5. Measure OD at 595 ± 20 nm. The signal is stable for up to 1 hour.

5. Data Analysis

Subtract the blank OD from the standard OD values and plot the OD against standard protein concentrations. Use the standard curve to determine the sample protein concentration.



For further technical questions please do not hesitate to contact us by email (technical@abcam.com) or phone (select “*contact us*” on www.abcam.com for the phone number for your region).

Technical Support

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