

Amplite™ Colorimetric Bradford Protein Quantitation Assay Kit

 Catalog number: 11118, 11119
 Unit size: 1000 Tests, 5000 Tests

Component	Storage	Amount (Cat No. 11118)	Amount (Cat No. 11119)
Component A: Bradford Assay Solution	Refrigerated (2-8 °C), Minimize light exposure	50 mL	250 mL
Component B: BSA Standard (1 mg/mL)	Freeze (< -15 °C), Minimize light exposure	1 mL	5 mL

OVERVIEW

The traditional Bradford protein assay is widely used for quantifying protein concentrations. However, many of the commercial protocols are complicated. Amplite™ Colorimetric Bradford Protein Quantitation Assay Kit is a two-component and detergent-compatible assay to determine total protein concentrations. The assay is based on the same Coomassie Blue G-250 protein indicator as Bradford protein assay and provides comparable accuracy. Our proprietary formulation makes our kit much more convenient and rapid. The protein signal is monitored around 600 nm and assay is completed within 30 minutes. Amplite™ Colorimetric Bradford Protein Quantitation Assay Kit can be performed in a convenient 96-well microtiter-plate format and easily adapted to automation with no separation steps required.

AT A GLANCE

Protocol summary

1. Prepare Bradford working solution (50 µL)
2. Add BSA standards or test samples (50 µL)
3. Incubate at room temperature for 5 - 15 minutes
4. Read absorbance at 595 nm

Important

Thaw all the kit components at room temperature before use.

KEY PARAMETERS

Absorbance microplate reader

Absorbance 595 nm
 Recommended plate Clear bottom

PREPARATION OF STANDARD SOLUTION

For convenience, use the Serial Dilution Planner:

<https://www.aatbio.com/tools/serial-dilution/11118>

BSA Standard

Add 20 µL of 1 mg/mL BSA Standard (Component C) to 480 µL of PBS (not provided) to generate 40 µg/mL BSA standard solution (BS1). Then perform 1:2 serial dilutions in PBS to get serially diluted BSA standards BS2 - BS7. Note: It is necessary to create a standard curve for each assay.

SAMPLE EXPERIMENTAL PROTOCOL

Table 1. Layout of BSA standards and test samples in a clear bottom 96-well microplate. BS= BSA Standards (BS1 - BS7, 40 to 0.625 µg/mL); BL=Blank Control; TS=Test Samples

BS1	BS1	TS	TS
BS2	BS2
BS3	BS3		
BS4	BS4		
BS5	BS5		
BS6	BS6		
BS7	BS7		
BL	BL		

Table 2. Reagent composition for each well

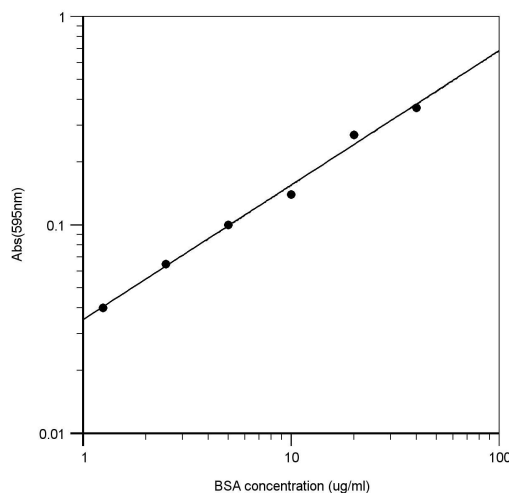
Well	Volume	Reagent
BS1-BS7	50 µL	Serial Dilutions (40-0.625 µg/mL)
BL	50 µL	PBS
TS	50 µL	Test Samples

1. Prepare BSA standards (BS), blank controls (BL), and test samples (TS) according to the layout provided in Tables 1 and 2.
2. Add 50 µL of Bradford working solution to each well of BSA standard, blank control, and test samples to make the total assay volume of 100 µL/well.
3. Incubate the reaction at room temperature for 5 to 15 minutes.
4. Read absorbance with an absorbance microplate reader at OD 595 nm.

EXAMPLE DATA ANALYSIS AND FIGURES

The reading (Abs(595nm)) obtained from the blank standard well is used as a negative control. Subtract this value from the other standards' readings to obtain the base-line corrected values. Then, plot the standards' readings to obtain a standard curve and equation. This equation can be used to calculate BSA concentration samples. We recommend using the Online Linear Regression Calculator which can be found at:

<https://www.aatbio.com/tools/linear-logarithmic-semi-log-regression-online-calculator>



BSA dose responses were measured with Amplite™ Colorimetric Bradford Protein Quantitation Assay Kit using a clear bottom 96-well plate.

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Figure 1. BSA dose responses were measured with Amplitude™ Colorimetric Bradford Protein Quantitation Assay Kit using a clear bottom 96-well plate.

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