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	18 mL	90 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 Braford 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 Braford 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 1X Bradford working solution (90 µL)
2. Add BSA standards or test samples (10 µL)
3. Incubate at room temperature for 5 - 15 minutes
4. Measure the absorbance at 595 nm and 460 nm, calculate the ratio of A595/A460 nm

Important

Thaw all the kit components at room temperature before use.

KEY PARAMETERS

Absorbance microplate reader

Absorbance	595 nm, 460 nm
Recommended plate	Clear bottom

PREPARATION OF STANDARD SOLUTIONS

For convenience, use the Serial Dilution Planner:
https://www.aatbio.com/tools/serial-dilution/11118

BSA Standard Solution
Add 50 µL of 1 mg/mL BSA Standard (Component C) to 50 µL of ddH2O (not provided) to generate 0.5 mg/mL BSA standard solution (BS7). Then perform 1:2 serial dilutions in ddH2O to get serially diluted BSA standards BS6 - BS1. Note: It is necessary to create a standard curve for each assay.

PREPARATION OF WORKING SOLUTION

Bradford working solution (1X)

Prepare 1X Bradford working solution by diluting 1 part Bradford Assay Solution (Component A) to 4 parts of ddH2O.

SAMPLE EXPERIMENTAL PROTOCOL

Table 1. Layout of BSA standards and test samples in a clear bottom 96-well microplate. BS= BSA Standards (BS1 - BS7, 0.5 to 0.0078 mg/mL); BL=Blank Control; TS=Test Samples

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

Table 2. Reagent composition for each well

Well	Volume	Reagent
BS1-BS7	10 µL	Serial Dilutions (0.5 to 0.0078 mg/mL)
BL	10 µL	PBS
TS	10 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 90 µL of 1X 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. Measure the absorbance with an absorbance microplate reader at OD 595 nm and 460 nm, and calculate the ratio of A595/A460 nm.

   Note: Please read 595 nm only if no 460 nm is available for the instrument.

EXAMPLE DATA ANALYSIS AND FIGURES

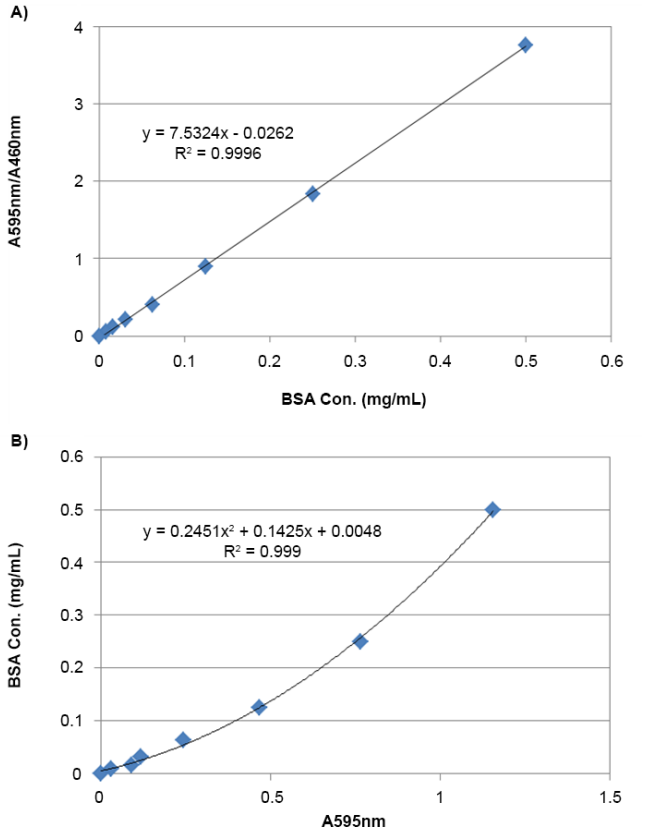

Figure 1. BSA dose responses were measured with Amplite® Colorimetric Bradford Protein Quantitation Assay Kit using a clear bottom 96-well plate. A) Detect with A595/A460nm. B) Detect with A595 nm if A460 nm is not available.

DISCLAIMER

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