



YamayBio

# Ready-to-use Bradford Protein Assay Kit (Detergent Compatible)

QUICK START GUIDE

For research use only.  
Not for use in diagnostic procedures.

# Contents and Storage

Bradford Protein Assay Kit (Detergent Compatible)

BH5485 800 Assays

**Storage:** Store at 4 °C /-20 °C for 12 months. The product is shipped with ice packs.

Component	Cat. No.	Volume	Storage
Bradford Reagent	BH5403	250 mL	4°C
BSA standards ① 0 µg/mL	BH5411	1 mL	-20°C
BSA standards ② 100 µg/mL	BH5412	1 mL	
BSA standards ③ 150 µg/mL	BH5414	1 mL	
BSA standards ④ 200 µg/mL	BH5415	1 mL	
BSA standards ⑤ 300 µg/mL	BH5417	1 mL	
BSA standards ⑥ 400 µg/mL	BH5418	1 mL	
BSA standards ⑦ 500 µg/mL	BH5419	1 mL	

## Introduction

The Ready-to-use Bradford Protein Assay Kit is a well-established method for protein quantification. Its principle is based on the binding of Coomassie Brilliant Blue dye to basic and aromatic amino acid residues in proteins, forming a stable blue complex. This complex has a maximum absorbance at 595 nm, and the absorbance is proportional to the protein concentration within a certain range.

This kit is a modified version of the traditional Bradford method, optimized for a detection range of **100–500 µg/mL**. The assay delivers rapid results within minutes and is compatible with samples containing high concentrations of reducing agents. It is also resistant to interference from commonly used detergents, such as Tween-20, 1% SDS, 1% NP-40, 1% Triton X-100, and 1% Brij-35. The kit includes pre-diluted BSA protein standards, eliminating the need for serial dilutions and simplifying the workflow.

# Quick Protocol

1. **10  $\mu\text{L}$**  sample or standards + **300  $\mu\text{L}$**  of Working Reagent.
2. Incubate for 3-5 minutes at room temperature.
3. Read at 595 nm.

## Detailed Procedures for BCA Protein Assay

1. Pipette **10  $\mu\text{L}$**  of each standard or unknown sample into the appropriate wells of a microplate (e.g., a 96-well plate).  
**Note:** It is strongly recommended to dilute samples with 1× PBS or 0.9% saline to ensure they fall within the working range.
2. Add **300  $\mu\text{L}$**  of Bradford Reagent to each well and mix thoroughly.
3. Incubate for 3–5 minutes at room temperature.
4. Measure the absorbance at or near 595 nm using a plate reader.
5. Subtract the average absorbance of the blank replicates from the 595 nm absorbance values of all standards and unknown samples.
6. Generate a standard curve by plotting the average blank-corrected absorbance at **595 nm** against the BSA standard concentrations ( $\mu\text{g/mL}$ ). Use this standard curve to determine the protein concentration of each unknown sample.

### Note:

1. When using curve-fitting software with a microplate reader, a four-parameter logistic (4PL) or best-fit curve typically provides more accurate results than a purely linear fit. When plotting data manually, a point-to-point curve is preferred over a straight line.
2. To calculate the original protein concentration of a diluted sample, multiply the measured concentration by the dilution factor.