

## Protein Estimation by BCA Method

#### **Product Overview**

Introducing DX/DT Enzymes' BCA Reagent A and B for precise protein estimation. These cutting-edge reagents ensure accuracy, sensitivity, and ease of use in molecular biology applications. Compatible with various sample types, they simplify high-throughput experiments while maintaining consistency and reproducibility. Manufactured under stringent quality control, DX/DT Enzymes guarantees reliable results, setting a new standard for protein quantification in research and diagnostics. Trust in the precision and quality of BCA Reagent A and B to advance your molecular biology experiments with confidence.

#### Storage

- BCA Reagent A and B, Room
  Temperature (15 to 30 ℃)
- Working Reagent (WR)
  (A+B), 4°C

#### Protocol

### Catalog

- RC20-1 > 100ml Reagent A,
  5ml Reagent B
- RC20-3 > 1000ml Reagent A,
  50 ml Reagent B

# PART 1: PREPARATION OF WORKING REAGENT (WR)

Prepare WR by mixing 50 parts of BCA Reagent A with 1 part of BCA Reagent B (50:1, Reagent A:B). For example, combine 100 mL of Reagent A with 2mL of Reagent B. Total volume of working reagent now is 102 mL. When reagents are uniformly mixed, a vibrant green colour shall appear. You can store the working reagent at 4°C for 7 days. It is recommended to prepare fresh working reagent based on number of unknown samples, everytime.

For One Sample, 2ml of working reagent is taken for the assay.



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# PART 2: PREPARATION OF WORKING STANDARD (WS)

BSA Stock Concentration: 2mg/ml

The BSA standard can be diluted based on expected protein concentration ranges. Diluent can be buffer/matrix in which your unknown protein of interest is currently present.

Example protein working range: 200µg - 1000µg/ml.

Example calculations are given below

Vial Name	Diluent (μL)	BSA Stock (μL)	Total (µL)	Final BSA concentration (µg/ml)
S1	90	10	100	200
S2	80	20	100	400
S3	70	30	100	600
S4	60	40	100	800
S5	50	50	100	1000
Unknown 1, 2, 3	3 0	0	100	?

If you suspect that, your unknown sample is concentrated, you can dilute it accordingly. Final volume of any sample MUST be 100µL.

### PART 3: STEP BY STEP PROCEDURE FOR THE ASSAY

- 1. Take 100µL of above samples in a clean glass test tube.
- 2.Add 2mL of working reagent to each tube and mix well.
- 3. Incubate the tube at selected temperature and time
- Standard Protocol: 37°C for 30 minutes (working range = 200–1000 μg/mL).
- RT Protocol: RT for 2 hours (working range = 200–1000 μg/mL)
- Enhanced Protocol: 60°C for 15-30 minutes (working range = 5-250 μg/mL)



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<u>Use a water bath to heat tubes for either Standard (37°C incubation) or Enhanced (60°C incubation) Protocol. Using a forced-air incubator can introduce significant error in color development because of uneven heat transfer.</u>

- 4. Cool all the test tubes
- 5. Set the spectrophotometer to read at <u>562nm wavelength</u>. ZERO the instrument, using diluent (BLANK SAMPLE)
- 6. Take out the reading for all the standard and unknown samples within 10 minutes or as fast as possible. Prolonging the reading time may interfere with absolute readings.
- 7.Prepare a standard curve by plotting 11 562 nm measurement for each BSA standard vs. its concentration in µg/mL. Use the standard curve to determine the protein concentration of each unknown sample.

#### Other related products

CDNA Synthesis KIT

LEO Prime mastermix

RIGEL -HIGH FIDELITY MASTERMIX

Onestep RTPCR kits

DNA Markers (100bp, 50bp and 1KB)

POLARIS SYBR GREEN Mastermix

POLARIS AMP Tagman probe mastermix

Common Buffers and Reagents

SAFESTAIN DYE

#### **Expert Services**

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Peptide Synthesis

**IVT** products

sgRNA, miRNA, Antisense RNA synthesis

NGS

**HYBRIDOMA** 

Protein Expression in Bacteria, Yeast,

Mammalin and Insects

# Ordering details

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