

PROTEIN CONCENTRATION DETERMINATION USING BCA PROCEDURE

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REAGENTS:

PIERCE/Fisher Scientific: BCA Protein assay kit, Product# 23226 or similar products.

BSA Standard Stock: 1ug/ul (**Note:** you can use the **NEB's** BSA control for restriction enzymes).

BCA Protein Assay Procedure

- 1) Add 2ul samples (e.g., from 100ul lysate of one well from 12- or 24-well plates) or BSA Standard (0ug, 1ug, 2ug, 3ug, 4ug, 8ug BSA) to 1.5ml sample tubes.
- 2) Mix 50 parts BCA reagent A with 1 part reagent B (**Note:** you will need 500ul per assay).
- 3) Add 500ul to each sample tube.
- 4) Mix and incubate for 15 minutes @ 60°C (**Note:** Incubation Temp and/or Time may vary, e.g., 37C to 60C and 15min to 30min, depending on protein concentrations. We recommend using 15min x 60C).
- 5) Cool down to RT.
- 6) Read OD @ 562nm, using "BCA reagent only" tube (i.e., no protein) as Blank, with disposable 1.0ml microcuvettes (**min. vol. >300ul**) (**Note:** You may also use ddH₂O tube for balance and set sample buffer tube as reagent control).

[Link to Hanli's Magic Protein Concentration Calculation Page](#)