ClearBand Bradford Reagent

500 ml

Cat No: BR05

Shipping : Ship at ambient temperature.
Storage : Stable for 1 year at 4°C.

General Information

ClearBand Bradford Reagent is formulated for a ready-to-use total protein analysis reagent used for quick measurement of total protein concentration. *ClearBand* Bradford Reagent contains Coomassie Brilliant Blue G-250, which associates with basic and aromatic amino acids, thus leading to a shift in absorbance during protein determination.

ClearBand Bradford Reagent offer an easy-to-use assay in either test tube or microplate format: mix protein sample with the assay reagent, incubate shortly and measure the absorbance at 595nm. Color response with Coomassie is non-linear with increasing protein concentration, therefore, a standard curve must be created with each assay.

Protocol (Standard Test Tube, Range: 25-1500 µg/ml)

1. Prepare a set of protein standards in the following concentrations with Bovine Serum Albumin (BSA, not provided) using the instructions provided in Table 1. As an initial step, dissolve 4 mg of BSA in 2 ml of ultrapure water. Refer this as Stock BSA that is in 2 mg/ml concentration. Use ultrapure water as diluent to prepare the standard set.

Table 1. Preparation of BSA Standards

Standard	Diluent	BSA	Final Concentration
1	0	300 µL Stock	2000 µg/ml
2	150 µL	450 µL Stock	1500 µg/ml
3	325 µL	325 µL Stock	1000 µg/ml
4	325 µL	325 µL Standard 2	750 µg/ml
5	325 µL	325 µL Standard 3	500 µg/ml
6	325 µL	325 µL Standard 5	250 µg/ml
7	325 µL	325 µL Standard 6	125 µg/ml
8	400 µL	100 µL Standard 7	25 µg/ml
9	400 µL	0 µL	Blank

2. Mix the *ClearBand* Bradford Reagent before use by gently inverting the bottle several times (Do not shake the bottle). Remove the amount of reagent needed and warm it to room temperature.

3. Add 20 µl of each standard and unknown samples into appropriately labeled test tubes at least in triplicates.

4. Add 1 ml of *ClearBand* Bradford Reagent to each tube and mix well.

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info@ecotechbiotech.com

5. Incubate samples for 10 minutes at room temperature.

6. Set the spectrophotometer to 595 nm. Zero the instrument on a cuvette filled only with water. Subsequently, measure the absorbance of all samples.

7. Subtract the average 595 nm measurement for the blank replicates from all other individual readings.

8. Prepare a standard curve by plotting the average blank-corrected 595nm measurement for each BSA standard vs. its concentration in µg/mL. Use the standard curve to determine the protein concentration of each unknown sample.

Protocol (Microplate, Range: 25-1500 µg/ml)

1. Prepare a set of protein standards in the following concentrations with Bovine Serum Albumin (BSA, not provided) using the instructions provided in Table 2. As an initial step, dissolve 4 mg of BSA in 2 ml of ultrapure water. Refer this as Stock BSA that is in 2 mg/ml concentration. Use ultrapure water as diluent to prepare the standard set.

Table 2. Preparation of BSA Standards

Standard	Diluent	BSA	Final Concentration
1	0	30 µL Stock	2000 µg/ml
2	15 µL	45 µL Stock	1500 µg/ml
3	32.5 µL	32.5 µL Stock	1000 µg/ml
4	32.5 µL	32.5 µL Standard 2	750 µg/ml
5	32.5 µL	32.5 µL Standard 3	500 µg/ml
6	32.5 µL	32.5 µL Standard 5	250 µg/ml
7	32.5 µL	32.5 µL Standard 6	125 µg/ml
8	40 µL	10 µL Standard 7	25 µg/ml
9	40 µL	0 µL	Blank

2. Mix the ***ClearBand*** Bradford Reagent before use by gently inverting the bottle several times (Do not shake the bottle). Remove the amount of reagent needed and warm it to room temperature.

3. Add 30 µl of each standard and unknown samples into appropriately labeled test tubes at least in triplicates.

4. Add 250 µl of ***ClearBand*** Bradford Reagent to each tube and mix thoroughly on plate shaker for 30 seconds.

5. Incubate samples for 10 minutes at room temperature.

6. Set the spectrophotometer to 595 nm. Zero the instrument on a cuvette filled only with water. Subsequently, measure the absorbance of all samples.

7. Subtract the average 595 nm measurement for the blank replicates from all other individual readings.

8. Prepare a standard curve by plotting the average blank-corrected 595nm measurement for each BSA standard vs. its concentration in µg/mL. Use the standard curve to determine the protein concentration of each unknown sample.

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