SOP: SP003

BCA Protein Assay

Materials and Reagents:
1. 10 ml BCA protein assay reagent A (Pierce catalog # 23223, note 1)
2. 200 μl BCA protein assay reagent B (Pierce catalog # 23224, note 1)
3. 0.1 mg/ml BSA standard
4. 0.2 mg/ml BSA standard
5. 0.6 mg/ml BSA standard
6. 1.0 mg/ml BSA standard
7. 1.4 mg/ml BSA standard
8. 1.6 mg/ml BSA standard
9. 2.0 mg/ml BSA standard
10. Sample to be assayed
11. 15 ml conical tube
12. 96 microtiter plate
13. 37°C incubator

Protocol:
1. _____ Add 10 μl of each standard to separate wells of the 96 well microtiter plate (note 2).
2. _____ Add 10 μl of the sample to separate wells of the 96 well microtiter plate (note 3).
3. _____ Add 10 μl of the blank to separate wells of the 96 well microtiter plate. This should be the same buffer that the sample is in.
4. _____ Mix the BCA protein assay reagents A and B together in a 15 ml conical tube.
5. _____ Add 200 μl of the above mixture to each well that contains sample, standard or blank.
6. _____ Incubate at 37°C for 30 minutes.
7. _____ Read the absorbance of the microtiter plate wells at 550 nm using the Microplate reader, and analyze the raw data using the curve fit option of the microplate reader software.

Notes:
1. The Pierce BCA assay reagents can also be purchased as a kit, catalog # 23225.
2. Each standard, sample and blank should be assayed in duplicate.
3. The samples should be assayed at multiple dilutions, such as undiluted, 1:2, 1:5, 1:10, and 1:100. All dilutions should be made with the buffer that the original sample is in.

References: