Product Manual

BCG Albumin Assay Kit

Catalog Number

MET-5017 250 assays

FOR RESEARCH USE ONLY
Not for use in diagnostic procedures

Introduction

Human serum albumin (HSA) is the most abundant protein in human blood plasma. Albumin constitutes about half of the blood serum protein. It is soluble and monomeric. The typical albumin concentration in blood is around 5 g/dL (50 mg/mL). It is produced in the liver as preproalbumin, which has an N-terminal peptide that is removed before the nascent protein is released from the rough endoplasmic reticulum. The product, proalbumin, is in turn cleaved in the Golgi vesicles to produce the secreted albumin. Albumin transports hormones, fatty acids, and other compounds, buffers pH, and maintains osmotic pressure.

Cell Biolabs' BCG Albumin Assay Kit is a simple, high-throughput colorimetric assay developed for the detection and quantitation of albumin in plasma, serum, urine or other biological fluid samples. The kit utilizes a proprietary BCG (Bromocresol Green) formulation, which forms a color complex specifically with albumin in samples (no pretreatment is required). The assay is performed in a 96-well microtiter plate format and measured within 5 minutes. Each kit provides sufficient reagents to perform up to 250 assays, including standards and unknown samples. The kit contains an albumin standard and has a detection sensitivity limit of ~ 0.02 g/dL.

Kit Components

1. Albumin Standard (Part No. 50171C): One 1 mL vial of 5 g/dL BSA (sterile filtered).
2. BCG Reagent (Part No. 50172B): One 50 mL amber bottle.

Materials Not Supplied

1. 96-well microtiter plate
2. 10 µL to 1000 µL adjustable single channel micropipettes with disposable tips
3. 50 µL to 300 µL adjustable multichannel micropipette with disposable tips
4. Multichannel micropipette reservoir
5. Microplate reader capable of reading in the 570-670 nm range (620 nm is optimal)

Storage

Upon receiving, aliquot and store the Albumin Standard at -20ºC to avoid multiple freeze/thaws. The BCG Reagent is light sensitive and should be stored at 4ºC.

Preparation of Reagents

- Components should be thawed/maintained at room temperature during assay preparation. Any unused Albumin Standard should be aliquoted and frozen at -20ºC to avoid multiple freeze/thaws.

Preparation of Albumin Standard

- Freshly prepare a dilution series of the standard in the concentration range of 0 g/dL – 5 g/dL by diluting the standard stock solution (provided at 5 g/dL) in deionized water (see Table 1). Albumin Standards should be prepared fresh.

Standard Tubes	Albumin Standard	Deionized Water	Final Albumin Standard Concentration (g/dL)
1	50 µL	0 µL	5.0
2	40 µL	10 µL	4.0
3	30 µL	20 µL	3.0
4	20 µL	30 µL	2.0
5	15 µL	35 µL	1.5
6	10 µL	40 µL	1.0
7	5 µL	45 µL	0.5
8	0 µL	50 µL	0

Table 1. Preparation of Albumin Standards

- Optional: If a higher sensitivity standard curve is desired, freshly prepare prepare a dilution series of the standard in the concentration range of 0.015 g/dL – 1.0 g/dL (1:2 dilution recommended) by diluting the standard stock solution in deionized water. Use standards immediately; do not store.

Preparation of Samples

- Plasma: Collect blood with an anticoagulant such as heparin, citrate or EDTA and mix by inversion. Centrifuge the blood at 1000 x g at 4°C for 10 minutes. Collect plasma supernatant without disturbing the white buffy layer. Sample should be tested immediately or frozen at -80°C for storage. Plasma must be diluted before assaying (1:2 in water).
- Serum: Collect blood in a tube with no anticoagulant. Allow the blood to clot at room temperature for 30 minutes. Centrifuge at 2500 x g for 20 minutes. Remove the yellow serum supernatant without disturbing the white buffy layer. Samples should be tested immediately or frozen at -80°C for storage. Serum must be diluted before assaying (1:2 in water).
- Urine: Collected urine samples should be stored at -80ºC prior to performing the assay. Urine samples with visible particulates should be centrifuged or filtered prior to testing. Urine should be assayed undiluted.

Assay Protocol

Samples should be assayed in duplicate or triplicate. A freshly prepared standard curve should be used each time the assay is performed.

1. Add 10 µL of the diluted Albumin Standard or sample to the 96-well microtiter plate.
2. Add 200 µL of BCG Reagent to each well.
3. Cover the plate wells to protect the reaction from light.
4. Incubate at room temperature for 5 minutes on an orbital shaker.
5. Read absorbance in the 570-670 nm range (620 nm is optimal).
6. Calculate the concentration of albumin within samples by comparing the sample absorbance to the standard curve. Negative controls (without albumin) should be subtracted.

Conversion Factors for Albumin

0.1 g/dL = 1 mg/mL = 15 µM = 0.1% = 1,000 ppm

Example of Results

The following figures demonstrate typical BCG Albumin Assay Kit results. One should use the data below for reference only. This data should not be used to interpret actual results.

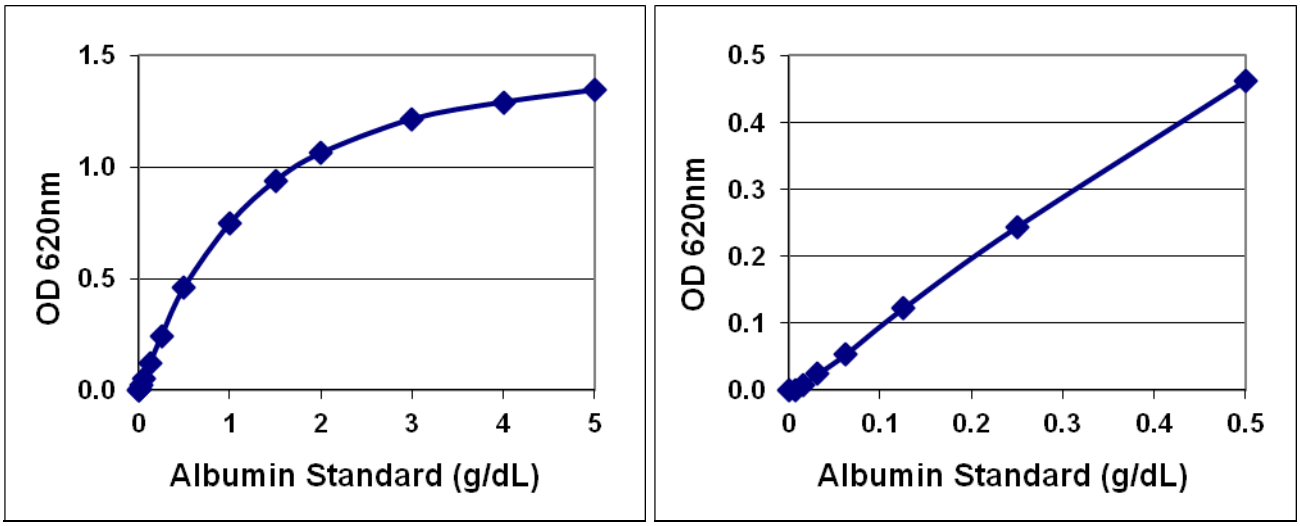

Figure 1: BCG Albumin Assay Standard Curve.

References

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5. Roche, M., Rondeau, P., Singh, N.R. (2008) *FEBS Letters* **582**, 1783-1787.

Recent Product Citation

Elje, E. et al. (2019). The comet assay applied to HepG2 liver spheroids. *Mutation Research/Genetic Toxicology and Environmental Mutagenesis*. doi:10.1016/j.mrgentox.2019.03.006.

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Contact Information

Cell Biolabs, Inc.
7758 Arjons Drive
San Diego, CA 92126
Worldwide: +1 858-271-6500
USA Toll-Free: 1-888-CBL-0505
E-mail: tech@cellbiolabs.com
www.cellbiolabs.com