BCG Albumin Assay Kit

Catalog Number KA1612

250 assays

Version: 02

Intended for research use only

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Introduction

Intended Use

Applications:

- ✓ Direct assays: albumin in serum, plasma, urine, biological preparations.
- ✓ Drug discovery/Pharmacology: effects of drugs on albumin metabolism.

Features:

- ✓ Sensitive and accurate: Use as little as 5 µL samples. Detection range 0.01 g/dL (1.5µM) to 5 g/dL (750µM) albumin in 96-well plate assay.
- ✓ Simple and high-throughput: The procedure involves addition of a single working reagent and incubation for 5 min. Can be readily automated as a high-throughput assay in 96-well plates for thousands of samples per day.
- ✓ Improved reagent stability and versatility: The optimized formulation has greatly enhanced reagent and signal stability. Cuvet or 96-well plate assay.
- ✓ No interference in biological samples. No pretreatments are needed. Assays can be directly performed on raw biological samples i.e., in the presence of lipid and protein.

Background

Albumin is the most abundant plasma protein in human. It accounts for about 60% of the total serum protein. Albumin plays important physiological roles, including maintenance of colloid osmotic pressure, binding of key substances such as long-chain fatty acids, bile acids, bilirubin, haematin, calcium and magnesium. It has anti-oxidant and anticoagulant effects, and also acts as a carrier for nutritional factors and drugs, as an effective plasma pH buffer. Serum albumin is a reliable prognostic indicator for morbidity and mortality, liver disease, nephritic syndrome, malnutrition and protein-losing enteropathies. High levels are associated with dehydration. Simple, direct and automation-ready procedures for measuring albumin concentration in biological samples are becoming popular in Research and Drug Discovery. BCG Albumin Assay Kit is designed to measure albumin directly in biological samples without any pretreatment. The improved method utilizes bromcresol green that forms a colored complex specifically with albumin. The intensity of the color, measured at 620nm, is directly proportional to the albumin concentration in the sample. The optimized formulation substantially reduces interference by substances in the raw samples.

General Information

Materials Supplied

List of component

Component	Amount
Reagent	50 mL
Albumin standard	1 mL 5 g/dL BSA

Storage Instruction

Store Reagent and standard at 4℃ and -20℃, respectively. Shelf life of 12 months after receipt.

Materials Required but Not Supplied

Pipeting devices and accessories (e.g. 5 µL).

- Procedure using 96-well plate:

Clear bottom 96-well plates (e.g. Corning Costar) and plate reader.

- Procedure using cuvette:

Spectrophotometer and cuvets for measuring OD at 620nm.

Precautions for Use

Precautions:

Reagents are for research use only. Normal precautions for laboratory reagents should be exercised while using the reagents.

Assay Protocol

Assay Procedure

- Reagent Preparation:

Important: bring reagent to room temperature and shake before use.

- Procedure using 96-well plate:

1. Dilute standards in distilled water as follows. Dilute serum samples 2 fold. Transfer 5 µL diluted standards and diluted samples to wells of a clear bottom plate. Store diluted standards at -20℃ for future use.

No	STD + H_2O	Vol (µL)	BSA (g/dL)
1	100µL + 0µL	100	5.0
2	80µL + 20µL	100	4.0
3	60µL + 40µL	100	3.0
4	40µL + 60µL	100	2.0
5	30µL + 70µL	100	1.5
6	20µL + 80µL	100	1.0
7	10µL + 90µL	100	0.5
8	0µL + 100µL	100	0

2. Add 200 µL working reagent and tap lightly to mix. Avoid bubble formation!
3. Incubate 5 min at room temperature and read optical density at 570-670nm (peak absorbance at 620nm).

- Procedure using cuvette:

1. Transfer 20 µL Blank, Standards and samples to appropriately labeled tubes. Add 1000 µL working reagent and tap lightly to mix. Incubate 5 min at room temperature.
2. Transfer to cuvet and read optical density at 620nm.

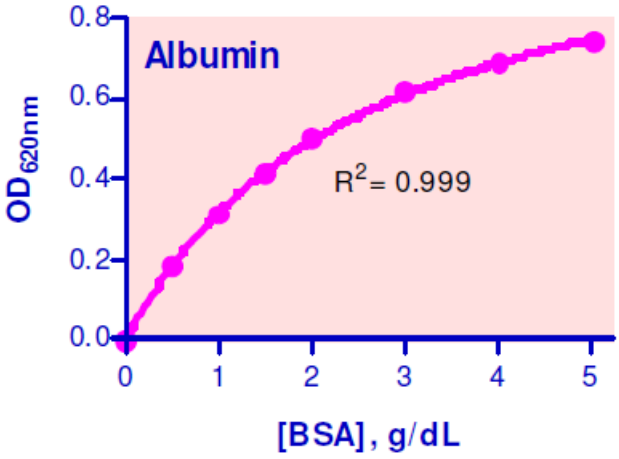
Important: if sample OD is higher than the OD for standard, dilute samples with distilled water and repeat the assay.

Data Analysis

Calculation of Results

Subtract blank OD (water, #8) from the standard OD values and plot the OD against standard concentrations. Use the standard curve to determine the sample albumin concentration.
Conversions: 0.1 g/dL albumin equals 15 µM, 0.1% or 1000 ppm.

Albumin was assayed in duplicate using the 96-well assay protocol. The albumin content (g/dL) was 4.8 ± 0.0 and 5.4 ± 0.0 in human serum and plasma, 2.2 ± 0.0 and 2.8 ± 0.2 in rat serum and plasma, 3.2 ± 0.2 in goat serum and 2.0 ± 0.0 in fetal bovine serum, respectively. Albumin in a fresh healthy human urine sample was below the detection limit (0.01 g/dL).



Standard Curve in 96-well plate assay

Resources

References

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- ✓ Cosgrove, D. et al (2008). Integrin alpha1β1 Regulates Matrix Metalloproteinases via P38 Mitogen-Activated Protein Kinase in Mesangial Cells. Implications for Alport Syndrome. Am. J. Pathology 172:761-773.