Albumin Assay Kit

Catalog No.: abx298891

Size: 96 tests

Detection Range: 0.08 g/L – 15 g/L

Sensitivity: 0.08 g/L

Storage: Store the Chromogenic Reagent at 4°C in the dark. Store the Standards at -20°C in the dark.

Application: For detection and quantification of Albumin content in serum, plasma, and cell culture supernatant.

Introduction

Abbexa's Albumin Assay Kit is a quick, convenient, and sensitive method for measuring and calculating Albumin content. Bromocresol Green dye combines with albumin at pH 4.0 – 4.2, forming a yellow-colored complex with an absorbance maximum at 630 nm. The intensity of the color is proportional to the Albumin content, which can then be calculated.

Kit components

1. 96-well microplate
2. Chromogenic Reagent: 6 ml
3. Standard (20 g/L): 2 × 1.2 ml
4. Plate sealer: 2

Materials required but not provided

1. Microplate reader (630 nm)
2. Double distilled water
3. Normal saline (0.9 % NaCl)
4. PBS (0.01 M, pH 7.4)
5. Pipette and pipette tips
6. 1.5 ml microcentrifuge tubes
7. Vortex mixer
8. Incubator

Protocol

A. Preparation of samples and reagents

1. Samples

Isolate the test samples soon after collecting and analyze immediately or aliquot and store at -80°C for up to one month. Avoid multiple freeze-thaw cycles.

- **Serum/Plasma:** Samples can be tested directly.
- **Cell Culture Supernatant:** Samples can be tested directly.

It is recommended to carry out a preliminary experiment to determine the optimal dilution factor of samples before carrying out the formal experiment. Dilute samples into different concentrations with normal saline (0.9 % NaCl) or PBS (0.01 M, pH 7.4), then carry out the assay procedure. The recommended dilution factor for different samples is as follows (for reference only):

Sample Type	Dilution Factor
Human serum	8 – 15
Human plasma	8 – 15
HepG2 supernatant	1
Mouse plasma	8 – 15
Rat serum	8 – 15

Note:

- Fresh samples or recently obtained samples are recommended to prevent degradation and denaturalization that may lead to erroneous results.

2. Reagents

- **Chromogenic Working Solution:** For each well to be used, mix 50 µl Chromogenic Reagent with 200 µl double distilled water. Prepare fresh before use.
- **Standards:** Take a standard vial from storage at -20°C and place on ice to thaw. Prepare diluted standards as summarized in the following table:

Standard Dilution (g/L)	0	1.0	2.0	3.5	5.0	8.0	12.0	15.0
20 g/L Standard (µl)	0	10	20	35	50	80	120	150
Double distilled water (µl)	200	190	180	165	150	120	80	50

Note:

- Allow all Chromogenic Reagent to equilibrate to room temperature before use.
- Keep Standard on ice during use, avoid repeated freeze-thaw cycles.

B. Assay Procedure

1. Add 10 µl of diluted standards to standard wells.
2. Add 10 µl of sample to sample wells.
3. Add 250 µl Chromogenic Working Solution to each well.
4. Incubate for 10 minutes at room temperature.
5. Measure the OD of each well with a microplate reader at 630 nm.

C. Calculation of Results

Plot the standard curve, using the average OD of duplicate standard dilution curves (adjusted for the blank) on the y-axis, and their respective concentrations on the x-axis. The curve should form a straight line, described by the formula $y = ax + b$. Based on this curve, the concentration of Albumin in each sample well can be derived with the following formulae:

1. Serum/Plasma/Cell Culture Supernatant samples:

$$\text{Albumin content (g/L)} = \frac{\Delta A_{630} - \boldsymbol{b}}{\boldsymbol{a}} \times \boldsymbol{f}$$

where:

ΔA_{630}	$OD_{Sample} - OD_{Blank}$
a	Gradient of the standard curve ($y = \boldsymbol{a}x + b$)
b	Y-intercept of the standard curve ($y = ax + \boldsymbol{b}$)
f	Dilution factor of the sample