DIATEK – ALBUMIN

Diagnostic reagent for quantitative in vitro determination of Albumin in human serum.

LINICAL SIGNIFICANCE

Albumin consists of approximately 60% of the total proteins in the body, the other major part being globulin. It is synthesized in the liver and maintains the osmotic pressure in blood. Albumin also helps in the transportation of drugs, hormones and enzymes.

INCREASES

Elevated levels are rarely seen and are usually associated with dehydration.

DECREASES

Decreased levels are seen in liver diseases (Hepatitis, Cirrhosis). Malnutrition, Kidney disorders, increased fluid loss during extensive burns and decreased absorption in gastro-intestinal diseases.

METHODOLOGY : BCG Method

PRINCIPLE

Albumin+Bromocresol Green → Green Albumin BCG Complex

REAGENT COMPOSITION

BCG Reagent

Succinate Buffer	-	90 mmol/L
Bromocresol Green	-	0.26 mmol/L
Standard Concentration	-	4.0 g/dl

STORAGE / STABILITY

1. BCG Reagent is stable at R.T. till the expiry mentioned on the label.
2. Albumin Standard is stable at 2-8^{0}C till the expiry mentioned on the label.

REAGENT PREPARATION

Reagents are ready to use. Protect from Bright light.

SAMPLE MATERIAL

Serum, EDTA Plasma, Albumin is reported to be stable in the sample for 6 days at 2-8^{0}C.

ASSAY PROCEDURE

Wavelength/Filter: 630 nm (Hg 623 nm) / Red
Temperature : R.T.
Light Path : 1 cm

Pipette into clean dry test tubes labeled as Blank (B), Standard (S) and Test (T)

Addition Sequence	B (ml)	S (ml)	T (ml)
BCG Reagent (A1)	1.0	1.0	1.0
Distilled water	0.01	-	-
Albumin Standard (S)	-	0.01	-
Sample	-	-	0.01

Mix well and incubate at R.T. for 5 min. Measure absorbance of the standard (Abs. S) and Test Sample (Abs. T) Against the Blank.

CALCULATIONS

$$\text{Albumin in g/dl} = \frac{\text{Abs. T}}{\text{Abs. S}} \times 4$$

Globulin in g/dl = (Total Proteins) – (Albumin)
(in g/dl) (in g/dl)

$$\text{A/G Ratio} = \frac{\text{Albumin in g/dl}}{\text{Globulin in g/dl}}$$

LINEARITY

The procedure is linear upto 8 g/dl. If values exceed this limit, dilute the sample with distilled water and repeat the assay. Calculate the value using the proper dilution factor.

NOTE

Gross haemolysis, ampicillin and heparin interfere with the results. Elevated bilirubin and lipemic samples may have a slight effect on accuracy. For grossly lipemic samples run a sample blank by adding 0.02 ml sample in 2 ml distilled water. Read the absorbance against D.W. and substract the blank absorbance.

QUALITY CONTROL

To ensure adequate quality each run should include assayed Normal and Abnormal controls

NORMAL REFERENCES VALUES

Serum, Plasma (Albumin): 3.7 – 5.3 g/dl
Globulin : 2.3 – 3.6 g/dl
A/g Ratio : 1.0 – 2.3

It is recommended that each laboratory establish it own normal range representing its patient population.

REFERENCES:

1. Doumas, B.T.Watson, W.A., (1971) Clin. Chem. Acta. 31:87

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DIATEK – TOTAL PROTEIN

Diagnostic reagent for quantitative in vitro determination of Total Protein in human serum.

CLINICAL SIGNIFICANCE

Proteins are constituents of muscle, enzymes, hormones and several other key functional and structural entities in the body. They are involved in the maintenance of the normal distribution of water between blood and the tissues. Consisting mainly of albumin and globulin the fractions vary independently and widely of diseases.

INCREASES

Increased levels are found mainly in dehydration.

DECREASES

Decreased levels are found mainly in malnutrition, impaired synthesis, protein losses as in hemorrhage or excessive protein catabolism.

PRINCIPLE

Protein+Cu^{++} ⟶ Blue Violet coloured complex

The intensity of the colour formed is directly proportional to the amount of total proteins present in the sample.

REAGENT COMPOSITION

Copper Sulphate	≥ 20 mmol/L
Potassium Sodium a Tartarate	≥ 15 mmol/L
Sodium Hydroxide	≥ 30 mmol/L
Surfactants	

STORAGE / STABILITY

Biuret Reagent is stable at R.T. till the expiry mentioned on the label.

Protein Standard is stable at 2-8^{0}C till the expiry mentioned on the label.

REAGENT PREPARATION

Reagents are ready to use. Protect from Bright light.

SAMPLE MATERIAL

Serum or Plasma. Proteins are reported to be stable in the sample for 6 days at 2-8^{0}C.

ASSAY PROCEDURE

Pipette into clean dry test tubes labeled as Blank (B), Standard (S) and Test (T)

Addition Sequence	B (ml)	S (ml)	T (ml)
Biuret Reagent (R_1)	1.0	1.0	1.0
Protein Standard (S)	-	0.01	-
Sample	-	-	0.01

Mix well and incubate at 37^{0}C for 5 mins. Or at R.T for 10 mins. Measure the absorbance of the Standard (Abs. S) and Test Sample (Abs.T) against the Blank, at 555 nm (520-570) within 60 Mins.

CALCULATIONS

$$\text{Total Protein in g/dl} = \frac{\text{Abs.T}}{\text{Abs.S}} \times 6$$

LINEARITY

The procedure is linear upto 15 g/dl. If values exceed this limit, dilute the sample with distilled water and repeat the assay. Calculate the value using the proper dilution factor.

NOTE

Do not use if the reagent shows turbidity or black precipitates.

QUALITY CONTROL

To ensure adequate quality each run should include assayed Normal and Abnormal controls

NORMAL VALUE

Serum and Plasma : 6.0-8.0 g/dl

It is recommended that each laboratory establish it own normal range representing its patient population.

REFERENCES:

1. Gornall, A.G., et al, (1949) Biol. Chem. 177:751
2. Doumas, B.T. (1975) Clin Chem. 21:1159

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