Albúmina

AA

Colorimetric method for the determination of albumin in serum

SUMMARY

Proteins are macromolecular organic compounds, widely distributed in the body.

Albumin is the main contributor of plasmatic total proteins. Within its multiple functions it can be mentioned:

- transport of a wide variety of substances like steroid hormones, fatty acids, bilirubin, catecholamines, which are insoluble in aqueous medium when in their free form, and
- maintenance of colloid osmotic pressure, as a result of its low molecular weight and its high net load.

Abnormal albumin increases are occasional and often related to dehydration which cause reduction in the content of plasmatic water.

Hypoalbuminemia occurs in pathologic conditions such as excessive loss of proteins in nephrotic syndrome, malnutrition, prolonged infections, and severe burns. Other causes are decrease in the albumin synthesis by a poor diet, hepatic disease or malabsorption.

PRINCIPLE

Albumin reacts specifically (without previous separation) with the anionic form of the 3,3',5,5'-tetrabromo cresolsulfon phthalein (BCG). Increase of absorbance at 625 nm with respect to the reagent Blank is proportional to the albumin concentration in the sample.

PROVIDED REAGENTS

A. Reagent A: 0.3 mmol/l BCG solution, 0.1 mol/l acetate buffer and 0.9 g/l polyoxyethylene lauryl ether.

NON-PROVIDED REAGENTS

Wiener lab.'s **Calibrador A plus / Proti 2 Suero Patrón**.

INSTRUCTIONS FOR USE

Provided Reagent: ready to use.

WARNINGS

The Reagent is for “in vitro” diagnostic use.

Use the reagents according to the working procedures for clinical laboratories.

The reagents and samples should be discarded according to the local regulations in force.

STABILITY AND STORAGE INSTRUCTIONS

Provided Reagent: stable at room temperature until the expiration date shown on the box.

SAMPLE

Serum

a) Collection: obtain serum free from hemolysis.

b) Additives: not required.

c) Known interfering substances: there is no interference from bilirubin up to 200 mg/l, triglycerides up to 9 g/l or hemoglobin up to 700 mg/dl. See Young, D.S. in References for effect of drugs on the present method.

d) Stability and storage instructions: if serum is not immediately assayed it can be stored up to 3 days in refrigerator (2-10ºC) or one week in freezer (-4ºC).

REQUIRED MATERIAL (non-provided)

- Spectrophotometer of photocolorimeter.
- Micropipettes and pipettes for measuring the stated volumes
- Tubes or spectrophotometric cuvettes.
- Watch or timer.

ASSAY CONDITIONS

- Wavelength: 625 nm in spectrophotometer or 620-650 nm in photocolorimeter with red filter
- Reaction temperature: 15-28ºC
- Reaction time: 10 minutes
- Sample volume: 10 ul
- Reagent A volume: 2.5 ml
- Final reaction volume: 2.51 ml

PROCEDURE

In three photocolorimeter test tubes labeled B (Blank), S (Standard) and U (Unknown), place:

	B	S	U
Calibrador / Suero Patrón	-	10 ul	-
Sample	-	-	10 ul
Reagent A	2.5 ml	2.5 ml	2.5 ml

Mix with a rod. Incubate tubes between 15ºC and 28ºC for 10 minutes. Read in spectrophotometer at 625 nm or in photocolorimeter with red filter (620-650 nm) setting instrument to zero O.D. with Reagent Blank.

STABILITY OF FINAL REACTION

Color is stable for 20 minutes, so absorbance should be read within that period.

CALCULATIONS

$$\text{Albumin (g/dl)} = U \times f \qquad f = \frac{\text{Albumin (g/dl)*}}{S}$$

*Albumin concentration in **Calibrador A plus** or **Suero Patrón**

$$\text{A/G ratio} = \frac{\text{Albumin (g/dl)}}{\text{Total Proteins (g/dl) - Alb. (g/dl)}}$$

QUALITY CONTROL METHOD

Each time the test is performed, analyze two levels of a quality control material (**Standatrol S-E 2 niveles**) with known albumin concentration.

REFERENCE VALUES

The following range of values was obtained from a healthy population from both sexes, with mixed diet and ages between 17 and 40 years old.
Albumin: 3.5 to 4.8 g/dl
A/G Ratio: 1.2 to 2.2
It is recommended that each laboratory establishes its own reference values.

SI SYSTEM UNITS CONVERSION

Albumin (g/dl) x 10 = Albumin (g/l)

PROCEDURE LIMITATIONS

See Known interfering substances under SAMPLE.

PERFORMANCE

The assays were performed in an Express Plus analyzer(*).
a) Reproducibility: precision studies were performed following the guidelines contained in NCCLS (National Committee on Clinical Laboratory Standards) EP5-A document.

Within Run (n = 20)

Level	S.D.	C.V.
3.47 g/dl	± 0.073 g/dl	2.10 %
2.70 g/dl	± 0.067 g/dl	2.48 %
5.23 g/dl	± 0.117 g/dl	2.23 %

Run to run (n = 20)

Level	S.D.	C.V.
3.43 g/dl	± 0.137 g/dl	3.99 %
2.80 g/dl	± 0.100 g/dl	3.57 %

b) Recovery: by adding known amounts of albumin to different samples, a recovery between 98 and 100% was obtained.
c) Detection limit: depends on the photometer and wavelength used. According to the required sensitivity for a ΔA minimum of 0.001, the minimum detectable change of concentration would be of 0.01 g/dl.
d) Linearity: reaction is linear up to 7 g/dl.
e) Correlation: albumin levels of 123 specimens were determined using the Wiener lab's **Albúmina AA** kit and a commercial kit based on same principle. The correlation coefficient was:
r = 0.9942, slope b = 0.9933 and intercept a = 0.0999

PARAMETERS FOR AUTOANALYZERS

For programming instructions consult the user manual of the analyzer in use. For calibration, it can be used Wiener lab.'s **Calibrador A plus**.

WIENER LAB. PROVIDES

- 6 x 60 ml (Cat. Nº 1009300).
- 6 x 60 ml (Cat. Nº 1009601).
- 6 x 120 ml (Cat. Nº 1690008).
- 8 x 50 ml (Cat. Nº 1009240).

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Symbols

The following symbols are used in the packaging for Wiener lab. diagnostic reagent kits.

CE — This product fulfills the requirements of the European Directive 98/79 EC for "in vitro" diagnostic medical devices

EC REP — Authorized representative in the European Community

IVD — "In vitro" diagnostic medical device

Σ — Contains sufficient for <n> tests

Use by

Temperature limitation (store at)

Do not freeze

Biological risks

Volume after reconstitution

Cont. — Contents

LOT — Batch code

Manufactured by:

Harmful

Corrosive / Caustic

Irritant

Consult instructions for use

Calibr. — Calibrator

CONTROL — Control

CONTROL + — Positive Control

CONTROL − — Negative Control

REF — Catalog number

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