K-*ASSAY*®

Prealbumin

For the Quantitative Determination of Human Prealbumin in Serum

Cat. No. KAI-053

INTENDED USE

For the quantitative determination of human prealbumin in serum by immunoturbidimetric assay. FOR *IN VITRO* DIAGNOSTIC USE.

INTRODUCTION AND SUMMARY

Prealbumin (thyroxin-binding prealbumin) transports thyroid hormones thyroxin (T_4) and triodothyronine (T_3). It also transports vitamin A in association with retinol-binding globulin.

Prealbumin levels are useful in the evaluation of several clinical conditions. Levels are decreased in most forms of acute and chronic hepatic disease. Prealbumin is a negative acute phase reactant with decreased levels associated with diseases involving inflammation or tissue necrosis.

Prealbumin has a circulation life of less than 2 days and is therefore a sensitive indicator of protein-calorie malnutrition.

Prealbumin has been measured using a variety of methods, including radioimmunoassay (RIA), radial diffusion, nephelometric, and enzyme-linked immunosorbent assay. The **K-*ASSAY*®** Prealbumin assay uses an immunoturbidimetric format.

PRINCIPLE OF TEST

The **K-*ASSAY*®** Prealbumin assay quantifies prealbumin based on immunoturbidimetric assay. The reagent uses a goat polyclonal antibody specific for human prealbumin.

The antibody binds to the prealbumin in the serum forming light scattering immune complexes which increase the turbidity of the sample. Since the increase in turbidity is proportional to the amount of prealbumin in the sample, the prealbumin concentration can be determined by measuring this increase in turbidity. The increase in turbidity is measured at 340 nm. Prealbumin in the sample is quantitatively determined. The **K-*ASSAY*®** Prealbumin can be run using a two reagent clinical chemistry analyzer. Six calibrators are prepared using the **K-*ASSAY*®** Prealbumin Calibrator. These calibrators are used for quantifying the levels of prealbumin present in the patient's serum sample.

KIT COMPOSITION

Reagents (Liquid Stable)

R1: Buffer Reagent 4 x 20 mL
Tris(hydroxymethyl)aminomethane

R2: Antiserum Reagent 2 x 8 mL
Anti-human prealbumin goat antiserum (20%)

WARNINGS AND PRECAUTIONS

FOR *IN VITRO* DIAGNOSTIC USE. Rx only.

Not to be used internally in humans or animals. Normal precautions exercised in handling laboratory reagents should be followed.

Do not mix or use reagents from one test kit with those from a different lot number.

Do not use reagents past their expiration date stated on each reagent container label.

Do not pipette by mouth. Avoid ingestion and contact with skin.

Reagents in this kit contain sodium azide as a preservative. Sodium azide may form explosive compounds in metal drain lines. When disposing of reagents through plumbing fixtures, flush with copious amounts of water. For further information, refer to "Decontamination of Laboratory Sink Drains to Remove Azide Salts," in the Manual Guide-Safety Management No. CDC-22 issued by the Centers for Disease Control, Atlanta, Georgia.

REAGENT PREPARATION

The reagents are ready to use. They do not need to be reconstituted.

STORAGE AND HANDLING

All reagents should be stored refrigerated (2-8°C). Return all reagents to 2-8°C promptly after use. Unopened reagents can be used for up to 18 months from the date of manufacture, as indicated by the expiration date on the package and bottle labels.

REAGENT STABILITY

Opened reagents can be used for 1 month if stored at 2-8°C. Discard reagents if they become contaminated. Evidence of cloudiness or particulate material in solution is cause to discard.

INSTRUMENT

Measurement of absorbance is to be made with an instrument able to accurately read absorbance at 340 and 700 nm. Refer to the instrument manual from the manufacturer regarding the following:

a) Use or function
b) Installation procedures and requirements
c) Principles of operation
d) Performance characteristics, operating instructions
e) Calibration procedures including materials and / or equipment to be used
f) Operational precautions, limitations, and hazards
g) Service and maintenance information

SPECIMEN COLLECTION AND PREPARATION

It is recommended that specimen collection be carried out in accordance with NCCLS document M29-T2. No method can offer complete assurance that human blood samples will not transmit infection. Therefore, all blood samples should be considered potentially infectious.

Serum is required for this assay. Do not use plasma or patient samples contaminated with heparin. Blood should be collected and the serum collected as soon as possible. Soon after the blood is drawn, it should be allowed to clot, centrifuged, and the serum separated from the clot to a plastic tube (not glass). Samples not tested within 72 hours should be frozen at -20°C. Avoid multiple freeze-thaws.

Use plastic tubes for storing the sample, do not use glass.

AUTOMATED ANALYZER APPLICATION

Suitable for two-reagent automated analyzers that use a multi-point calibration method.

PROCEDURE

Materials Supplied

Reagent 1 (R-1) Buffer Reagent 4 x 20 mL
Reagent 2 (R-2) Antiserum Reagent 2 x 8 mL

Materials Required But Not Supplied

Calibrators: **K-*ASSAY*®** Prealbumin Calibrator, Cat. No. KAI-054C. (6 calibrators containing human serum with known levels of prealbumin).

Two Reagent Clinical Chemistry Analyzer:
Capable of accurate absorbance readings at 340 / 700 nm
Capable of accurately dispensing the required volumes
Capable of maintaining 37°C

Assay Procedure

Note: Allow all reagents and specimens to warm to room temperature. Mix all reagents gently before using.

An example of automated application (Hitachi 717):

Sample 5 µL
↓
• ← R1 (Buffer Reagent) 250 µL
↓ 37 °C, 5 min.
• ← R2 (Antiserum Reagent) 50 µL
↓ 37 °C, 5 min.
2-point endpoint, 340/700 nm

Automated Method (Example)

Chemistry Parameters for Automatic Analyzer

INSTRUMENT	Roche / Hitachi 717
TEMPERATURE	37°C
TEST	(PALB)
ASSAY CODE	(2 POINT) : (24) - (50)
SAMPLE VOLUME	(5) ( )
R-1 VOLUME	(250) ( ) (NO)
R-2 VOLUME	(50) ( ) (NO)
WAVELENGTH	(700) (340)
CALIB. METHOD	(NONLINEAR) (1) (6)
STD.(1) Conc.-POS.	(*1) - (1)
STD.(2) Conc.-POS.	(*2) - (2)
STD.(3) Conc.-POS.	(*3) - (3)
STD.(4) Conc.-POS.	(*4) - (4)
STD.(5) Conc.-POS.	(*5) - (5)
STD.(6) Conc.-POS.	(*6) - (6)
SD LIMIT	(999)
DUPLICATE LIMIT	(10000)
SENSITIVITY LIMIT	(0)
ABS. LIMIT (SLOPE)	(32000) (INCREASE)
PROZONE LIMIT	(-320000) (LOWER)
EXPECTED VALUE	(-99999) (99999)
PANIC VALUE	(-99999) (99999)
INSTRUMENT FACTOR	(1.00)

*1-6: Input concentration of calibrators.

Parameters for other automated analyzers are available.

CALIBRATION

It is recommended that prealbumin levels be determined using a multi-point calibration curve prepared using the **K-*ASSAY*®** Prealbumin Calibrator. It is recommended that the user determine calibration curve frequency as this depends on the instrument and type/number of other assays being performed. Initially, calibration should be performed each day.

QUALITY CONTROL

Normal and abnormal controls of known concentration should be included in each assay performed. These controls should fall within the stated values assigned to the controls. The validity of the assay is in question if the value for the controls generated by the assay's calibration curve does not fall within the stated range. Recalibrate if the value determined for the controls falls outside the stated recovery range.

LIMITATIONS OF PROCEDURE

The measurable range for prealbumin is between 0 to 60 mg/dL. Grossly lipemic samples and samples with very high triglyceride concentrations should be diluted 1 part sample with 1 part isotonic saline or filtered to decrease nonspecific light scattering. Multiply results by 2 to compensate for the dilution.

If the prealbumin concentration of a patient sample is greater than the highest calibrator value, dilute 1 part sample with 4 parts isotonic saline and reassay. Multiply results by 5 to compensate for the dilution.

This assay should not be used with plasma samples or with patient samples contaminated with heparin.

PERFORMANCE

Precision

The precision for the **K-*ASSAY*®** Prealbumin assay was determined using packaged reagents, pooled human serum, and a Hitachi 717 analyzer.

Precision Assay: Within Run

Sample I	Sample II	Sample III
N = 20	N = 20	N = 20
Mean = 11.2	Mean = 24.9	Mean = 57.0
SD = 0.145	SD = 0.289	SD = 0.767
CV = 1.29%	CV = 1.16%	CV = 1.35%

Accuracy / Correlation

A comparison of the **K-*ASSAY*®** Prealbumin assay and a similar Prealbumin assay was performed using a Hitachi 717. The test results provided the following data:

y = 0.9164x - 0.332
r = 0.9924
n = 50
x = Similar Prealbumin assay
y = **K-*ASSAY*®** Prealbumin assay

x min	= 5.7	y min	= 4.2
max	= 43.2	max	= 38.4
mean	= 22.89	mean	= 20.64

Linearity

Linearity tests were performed with dilutions of normal human serum spiked with prealbumin. Testing was linear from 0 to 60 mg/dL of prealbumin.

INTERFERENCE

Bilirubin C	No interference up to 20 mg/dL
Bilirubin F	No interference up to 20 mg/dL
Hemoglobin	No interference up to 470 mg/dL
Intralipid	No interference up to 500 mg/dL

EXPECTED VALUE

The expected value as reported in the scientific literature is between 16 to 40 mg/dL. Each laboratory should establish its own expected values using this kit.

REFERENCES

1. Peterson, P.A. "Studies on interaction between prealbumin, retinol-binding protein, and vitamin A," J. Biol. Chem., 246:44 (1971).
2. Oppenheimer, J.H., "role of plasma proteins in the binding, distribution, and metabolism of thyroid hormones," N. Engl. J. Med. 278:1153 (1968).
3. Killingsworth, L.M. and Savory, J., "Nephelometric Studies on the Precipitin Reactions," J. Clin. Chem., 19: 403-407, 1973.
4. Sternberg, J.C. " A Rate Nephelometer for Measuring Specific Proteins by Immunoprecipitin Reaction," Clin. Chem., 23:1456-64, 1977.

LABELING SYMBOLS

LOT	Lot Number
RGT	Reagent
	Expiration or “Use By” Date
REF	Catalog Number
IVD	For *In Vitro* Diagnostics Use
2-8 °C	Temperature Limitation. Store between 2 and 8 degrees C
	Manufacturer
	Consult Package Insert for Instructions for Use
EC REP	Authorized Representative in the European Community

EU AUTHORIZED REPRESENTATIVE

CE

EC REP

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