TRUEchemie Total Proteins Test Kit (BIURET)

for the quantitative determination of Total proteins concentration in human serum

INTENDED USE

The TRUEchemie Total Proteins Test Kit (BIURET) is used for the quantitative determination of total proteins concentration in human serum or plasma.

INTRODUCTION

Serum protein is involved in the maintenance of normal distribution of water between blood and tissues through osmotic pressure. Low protein is primarily caused by malnutrition, impaired synthesis, loss as by hemorrhage or excessive protein catabolism. Elevated protein levels are caused mainly by dehydration.

The determination of total protein in serum makes use of the Biuret color reaction, known since 1878. Past attempts to stabilize the cupric ions in the alkaline reagent were unsuccessful until the addition of sodium potassium tartrate as a complexing agent.

PRINCIPLE

$$\text{Protein} + Cu^{++} \xrightarrow[\text{pH}]{\text{Alkaline}} \text{Cu - Protein Complex}$$

Protein in serum forms a violet coloured complex when reacted with cupric ions in an alkaline solution. The intensity of the violet colour is proportional to the amount of protein present when compared to a solution with known protein concentration.

PACK SIZE

Kit size	**2 x 50 ml**
Cat. no.	**ADX281**
Kit contents	
1) Total Proteins Reagent	2 x 50 ml
2) Total Proteins Standard (6 g/dL)	1 x 5 ml

REAGENT COMPOSITION

1) Total Proteins Reagent:
Sodium hydroxide : 6 mmol/L
Copper sulfate : 12 mmol/L
Sodium potassium tartrate : 32 mmol/L
Non-reactive ingredients.
2) Total Proteins Standard : 6 g/dL

REAGENT PREPARATION

Ready to use reagents.

WARNINGS AND PRECAUTIONS

1. For *in vitro* diagnostic use.
2. Handle in accordance with good laboratory procedures. Avoid ingestion and eye or skin contact.
3. Specimens should be considered infectious and handled appropriately.
4. The disposal of the residues has to be done as per local legal regulations

CALIBRATION

The procedures are calibrated with the standard solution which is included with each series of tests. Its absorbance is used to calculate the results.

REAGENT STORAGE & STABILITY

The unopened reagents are stable till the expiry date stated on the bottle and kit label when stored at 2-8°C. Do not use reagents over the expiration date.

SPECIMEN COLLECTION AND STORAGE

Serum free from haemolysis. The sample will remain stable for up to 5 days when kept at 2 - 8 °C.

MATERIALS REQUIRED BUT NOT PROVIDED

1. Pipettes to accurately measure required volumes
2. Test tubes/rack
3. Timer
4. 37 °C heating block or water bath
5. Photometer capable of accurately measuring absorbance at 545 nm

TEST PROCEDURE

Wavelength : 545 nm
Temperature : 37°C
Prewarm the reagent to reaction temperature.

	Blank (µL)	Standard (µL)	Sample (µL)
Total Proteins Reagent	1000	1000	1000
Total Proteins Standard	--	10	--
Sample	--	--	10

Mix well and incubate for 3 min at 37ºC or 5 min at room temperature (20 - 25ºC). After incubation, zero the Photometer with the reagent blank. Read and record the incuated standard and samples.

Final colour stability: A minimum of 3 hours

$$\text{Calculation: } \frac{\text{Sample OD}}{\text{Standard OD}} \times 6 = \text{g total proteins / dL}$$

S.I. Units (g/dl) x 10 = mmol/L

QUALITY CONTROL

Quality Controls are recommended to monitor the performance of automated assay procedures. Each laboratory should establish its own Quality Control scheme and corrective actions if controls do not meet the acceptable tolerances.

EXPECTED VALUE

Normal values Serum : 6.6 - 8.7 g/dL
It is strongly recommended that each laboratory establish its own normal range.

PERFORMANCE CHARACTERISTICS

Sensitivity: 0.15 g/dL
Linearity: Upto 14 g/dL under the described assay conditions. Samples that have Total proteins values greater than 14 g/dL should be diluted with saline water (NaCl 0.9 %) 1:1, re-assayed and the results multiplied by 2. The linearity limit depends on the sample / reagent ratio, as well as the analyzers used.

PRECISION:

Intra-assay precision within run (n=10)	**Mean (g/dL)**	**SD (g/dL)**	**CV (%)**
Control Level - 1	6.7	0.1	1.6
Control Level - 2	4.6	0.1	2.2

Inter-assay precision run to run (n=12)	**Mean (g/dL)**	**SD (g/dL)**	**CV (%)**
Control Level - 1	6.7	0.1	1.2
Control Level - 2	4.4	0.1	2.8

The reagent was tested for 12 days, using two different Total Proteins concentrations. The coefficient of variation was <5%.

AUTOMATED PROCEDURE

Appropriate program sheet is available for different analyzers upon request.

METHOD COMPARISON

Results obtained using TRUEchemie Total Proteins reagent (y) did not show systematic differences when compared with another commercial reagent (x) with similar characteristics. The results obtained is below: The correlation coefficient (r^2) was 0.974 and the regression equation is y=0.877x+0.933. The results of the performance characteristics depend on the analyzer used.

INTERFERENCES

- Lipemia (intralipid) may affect the results.
- Bilirubin (20 mg/dl) does not interfere.
- Hemoglobin may affect the results.
- Other drugs and substances may interfere.
- Dextrans used as plasma volume expanders for the treatment of low blood pressure,- complex with copper and tartrate forming a precipitate

WASTE MANAGEMENT

Please refer to local regulation requirements.

SYSTEM PARAMETERS

Mode	:	End point
Std. conc.	:	6
Wave length	:	545 nm
Units	:	g/dL
Flow cell temp.	:	37°C
Blank	:	Reagent
Reagent volume	:	1000 µL
Sample volume	:	10 µL
Incubation	:	3 min. at 37°C
Low normal	:	6.6
High normal	:	8.7
Sensitivity	:	0.15
Linearity	:	14
Reaction Slope	:	Increasing

REFERENCES

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6. Henry. R.J., et al. Clinical Chemistry Principles and Technique s. Harper & Row, N.Y., 415 (1974).
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8. Tietz. N.W., Fundamentals of Clinical Chemistry. W.B. Saunders. Philadelphia, PA 299, (1976).
9. ISO 15223-1:2021 Medical devices — Symbols to be used with information to be supplied by the manufacturer — Part 1: General requirements

Index of Symbols

Consult instructions for use	REF Catalogue number	Caution
IVD *In vitro* diagnostic medical device	LOT Batch code	Non-sterile
2°C 8°C Temperature limit 2-8 °C	Do not re-use	Use-by date
Manufacturer	Date of manufacture	Keep dry
Do not use if package is damaged	Keep away from sunlight	

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