Total protein FS*

Diagnostic reagent for quantitative in vitro determination of total protein in serum or plasma on photometric systems

Order Information

Cat. No.	Kit size						
1 2311 99 10 021	R1	4 x	20 mL	+ R2	1 x	20 mL	
	+	1 x	3 mL	Standard			
1 2311 99 10 026	R1	5 x	80 mL	+ R2	1 x	100 mL	
1 2311 99 10 023	R1	1 x	800 mL	+ R2	1 x	200 mL	
1 2311 99 10 704	R1	8 x	50 mL	+ R2	8 x	12.5 mL	
1 2311 99 10 917	R1	8 x	60 mL	+ R2	8 x	15 mL	
1 2311 99 90 314	R1	10 x	20 mL	+ R2	2 x	30 mL	
1 2300 99 10 030		6 x	3 mL	Standard			

Summary [1,2]

Measurement of total protein is a useful test in a variety of disorders. Decreased total protein concentrations can be detected in defective protein synthesis in the liver, protein loss due to impaired kidney function, intestinal malabsorption or nutritional deficiency. Elevated protein levels occur in chronic inflammatory disorders, liver cirrhosis and dehydration.

Method

Photometric test according to biuret method

Principle

Proteins form a violet blue color complex with copper ions in alkaline solution. The absorbance of the color is directly proportional to the concentration.

Reagents

Components and Concentrations

R1:	Sodium hydroxide	100 mmol/L
	Potassium sodium tartrate	17 mmol/L
R2:	Sodium hydroxide	500 mmol/L
	Potassium sodium tartrate	80 mmol/L
	Potassium iodide	75 mmol/L
	Copper sulphate	30 mmol/L
Standard:		5 g/dL

Contains bovine serum albumin (< 5%)

Storage Instructions and Reagent Stability

The reagents and the standard are stable up to the end of the indicated month of expiry, if stored at 2 – 25°C and contamination is avoided. Do not freeze the reagents and protect them from light!

Warnings and Precautions

1. Reagent 1: Warning. H290 May be corrosive to metals. P234 Keep only in original container. P390 Absorb spillage to prevent material damage.
2. Reagent 2: Warning. H290 May be corrosive to metals. H315 Causes skin irritation. H319 Causes serious eye irritation. H412 Harmful to aquatic life with long lasting effects. P234 Keep only in original container. P264 Wash hands and face thoroughly after handling. P273 Avoid release to the environment. P280 Wear protective gloves/protective clothing/eye protection/face protection. P332+P313 If skin irritation occurs: Get medical advice/attention. P305+P351+P338 If in eyes: Rinse cautiously with water for several minutes. Remove contact lenses, if present and easy to do. Continue rinsing. P337+P313 If eye irritation persists: Get medical advice/attention.
3. The reagents contain sodium hydroxide. Do not swallow! If the reagents get in contact with skin or mucous membranes rinse immediately with water!
4. Total Protein Standard FS contains animal material. The standard should be handled as potentially infectious and with the same precautions used for patient specimens.
5. In serum or plasma of patients who have received large intravenous amounts of polydextrans, too high values can be measured with the biuret method. In such cases an alternative method (e.g. Kjeldahl) has to be used.
6. In very rare cases, samples of patients with gammopathy might give falsified results [5].
7. Please refer to the safety data sheets and take the necessary precautions for the use of laboratory reagents. For diagnostic purposes, the results should always be assessed with the patient's medical history, clinical examinations and other findings.
8. For professional use only!

Waste Management

Please refer to local legal requirements.

Reagent Preparation

The standard is ready to use.

Substrate Start

The reagents are ready to use.

Sample Start

Mix 4 parts of R1 with 1 part of R2
(e.g. 20 mL R1 + 5 mL R2) = mono reagent
Stability after mixing: 1 year at 2 – 25 °C

Materials required but not provided

NaCl solution 9 g/L
General laboratory equipment

Specimen

Serum or plasma

Stability [3]:	6 days	at	20 – 25°C
	4 weeks	at	4 – 8°C
	at least one year	at	–20°C

Freeze only once!
Discard contaminated specimens!

Assay Procedure

Application sheets for automated systems are available on request.

Wavelength	540 nm, Hg 546 nm
Optical path	1 cm
Temperature	20 – 25°C/37°C
Measurement	Against reagent blank

Substrate start

	Blank	Sample or standard
Sample or standard	-	20 µL
Dist. water	20 µL	-
Reagent 1	1000 µL	1000 µL
Mix, read absorbance A1 after 1 – 5 min. at 20 – 25 °C/ 37 °C, then add:		
Reagent 2	250 µL	250 µL
Mix, incubate for 5 min. at 20 – 25°C/37°C and read absorbance A2 within 60 min.		

ΔA = (A2 – A1) sample or standard

* fluid stable

Sample start

	Blank	Sample or standard
Sample or standard	-	20 µL
Dist. water	20 µL	-
Mono reagent	1000 µL	1000 µL
Mix, incubate for 5 min. at 20 – 25°C/37°C and read absorbance against the reagent blank within 60 min.		

ΔA = A Sample/Standard

Calculation

With standard or calibrator

$$\text{Total protein [g/dL]} = \frac{\Delta A \text{ Sample}}{\Delta A \text{ Std. / Cal.}} \times \text{Conc. Std / Cal. [g/dL]}$$

Calibrators and Controls

For the calibration of automated photometric systems, DiaSys TruCal U calibrator is recommended. The assigned values of the calibrator are traceable to the biuret method. DiaSys TruLab N and P controls should be assayed for internal quality control. Each laboratory should establish corrective action in case of deviations in control recovery.

	Cat. No.	Kit size
TruCal U	5 9100 99 10 063	20 X 3 mL
	5 9100 99 10 064	6 x 3 mL
TruLab N	5 9000 99 10 062	20 x 5 mL
	5 9000 99 10 061	6 x 5 mL
TruLab P	5 9050 99 10 062	20 x 5 mL
	5 9050 99 10 061	6 x 5 mL

Performance Characteristics

Measuring range

The test has been developed to determine total protein concentrations within a measuring range from 0.05 – 15 g/dL. When values exceed this range samples should be diluted 1 + 1 with NaCl solution (9 g/L) and the result multiplied by 2.

Specificity/Interferences

No interference was observed by ascorbic acid up to 30 mg/dL, bilirubin up to 40 mg/dL, hemoglobin up to 500 mg/dL, dextran up to 2000 mg/dL and lipemia up to 1000 mg/dL triglycerides. For further information on interfering substances refer to Young DS [4].

Sensitivity/Limit of Detection

The lower limit of detection is 0.05 g/dL.

Precision (at 37°C)

Intra-assay n = 20	Mean [g/dL]	SD [g/dL]	CV [%]
Sample 1	5.27	0.05	0.91
Sample 2	7.05	0.07	1.01
Sample 3	10.4	0.08	0.80

Inter-assay n = 20	Mean [g/dL]	SD [g/dL]	CV [%]
Sample 1	5.24	0.06	1.06
Sample 2	7.07	0.11	1.53
Sample 3	10.4	0.14	1.32

Method Comparison

A comparison of DiaSys Total protein FS (y) with a commercially available test (x) using 68 samples gave following results:
y = 1.00 x - 0.07 g/dL; r = 0.997

Reference Range [1]

	[g/dL]	
Adults:	6.6 – 8.8	
Children	Female	Male
1 - 30 day(s)	4.2 – 6.2	4.1 – 6.3
1 – 6 month(s)	4.4 – 6.6	4.7 – 6.7
6 months – 1 year	5.6 – 7.9	5.5 – 7.0
1 – 18 year(s)	5.7 – 8.0	5.7 – 8.0

Each laboratory should check if the reference ranges are transferable to its own patient population and determine own reference ranges if necessary.

Literature

1. Thomas L. Clinical Laboratory Diagnostics. 1st ed. Frankfurt: TH-Books Verlagsgesellschaft; 1998. p. 644-7.
2. Johnson Am, Rohlfs EM, Silverman LM. Proteins. In: Burtis CA, Ashwood ER, editors. Tietz Textbook of Clinical Chemistry. 3rd ed. Philadelphia: W.B Saunders Company; 1999. p. 477-540.
3. Guder WG, Zawta B et al. The Quality of Diagnostic Samples. 1st ed. Darmstadt: GIT Verlag; 2001; p. 42-3.
4. Young DS. Effects of Drugs on Clinical Laboratory Tests. 5th ed. Volume 1 and 2. Washington, DC: The American Assocation for Clinical Chemistry Press 2000.
5. Bakker AJ, Mücke M. Gammopathy interference in clinical chemistry assays: mechanisms, detection and prevention. ClinChemLabMed 2007;45(9):1240-1243.

Manufacturer

IVD CE

DiaSys Diagnostic Systems GmbH
Alte Strasse 9 65558 Holzheim Germany