EN

Total Protein (Biuret)

REF 984328

❶ 3 x 20 ml Reagent

INTENDED USE
Reagent for photometric determination of Total Protein in homogenous liquid samples using automated Thermo Scientific™ Arena™ or Gallery™ analyzer.

METHOD
Colorimetric test by cupric ions.
Method is performed at 37 °C, using 540 nm filter and for side wavelength 700 nm filter .

PRINCIPLE OF THE PROCEDURE
Protein forms a coloured complex with cupric ions in alkaline solutions (3). The formation of the complex is measured at 540 nm. The method employs EDTA as a chelating and stabilizing agent for cupric ions.

REAGENT INFORMATION		Barcode ID
Reagent	3 x 20 ml	736

Note: Labels of reagent vials have two barcodes.
For Arena analyzers, turn the short barcode to the barcode reader.
For Gallery analyzers, turn the long barcode to the barcode reader.

Concentrations

NaOH	1.0 mol/l
$CuSO_4$	9 mmol/l
KI	9 mmol/l
Na_2-EDTA	28 mmol/l

Precautions
Reagent is hazardous.
See separate sheet inside the kit for Hazardous- and Precautions-phrases: H290, H314, P280, P303 + P361 + P353, P305 + P351 + P338, P310
Exercise the normal precautions required for handling all laboratory reagents.
The product has to be disposed of as laboratory chemical in accordance with local regulations.

Preparation
The reagent R1 is ready-to-use.
Note: Check that there are no bubbles on the surface of the reagent when you insert vials into the analyzer.

Storage and Stability
Reagents in unopened vials are stable at 2…25 °C until the expiry date printed on the label. Do not freeze the reagents or expose to light.
Refer to the Application Notes of your analyzer for the on board stability of reagents.

SAMPLES

Sample Type
Food, beverage and other sample material.

Sample concentration and Arena/Gallery application
All method related details are in the separate application note.

If the Arena or Gallery applications have a primary dilution of 1+9, this means that every sample is automatically first diluted with 1+9.

Sample preparation
Beer samples can be used directly.
If the sample has substances interfering the measurement, please handle it according to the following suitable preparation procedure:

- Use clear, colorless and practically neutral liquid samples directly.
- Filter or centrifuge turbid solutions.
- Degas samples containing carbon dioxide.
- Crush or homogenize solid or semi-solid samples.
- Adjust acidic samples to pH 8 by adding sodium or potassium hydroxide solution and incubate for approx. 15 min.

TEST PROCEDURE
See a separate application for the Arena or Gallery analyzer.

Materials required but not provided
Distilled water (aseptic and free of heavy metals) and general laboratory equipment.

All material needed for calibration standard and QC samples.

Calibration
Water based calibration standard solution can be used.
As an example, make a stock of Albumin (from bovine serum, 1000772605, A7906-10G, Sigma-Aldrich, >99 %) to 10 g/l and use this as a standard for automated calibration.

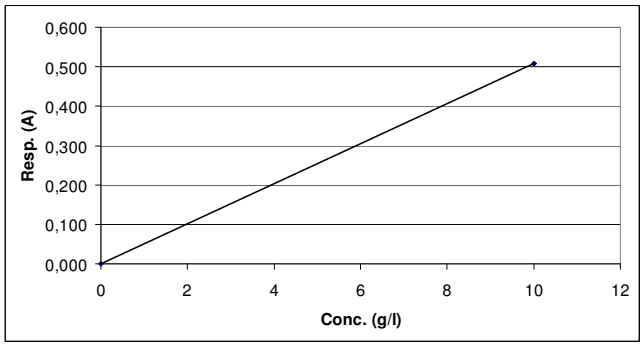
If the sample matrices differ significantly from the albumin, calibration can be performed, e.g. with wheat Glutein. This change should be validated by the user.

Quality Control
Use quality control samples at least once a day and after each calibration and every time a new bottle of reagent is used. It is recommended to use two level of controls. The control intervals and limits must be adapted to the individual laboratory requirements. The results of the quality control sample(s) should fall within the limits pre-set by the laboratory.

Quality control sample should be prepared from other material than calibration stock standard. It is recommended to set the value for the QC sample also with a reference method and prepare it from the same matrices as the samples. f the QC samples made from matrices similar to the samples gives too low or high results, this matrix effect should be removed by using the similar matrices for calibration standards as well.

CALCULATION OF RESULTS
The results are calculated automatically by the analyzer using a calibration curve.

Calibration Curve (example)



Calibrator	Response (A)	Calc. conc. (g/l)
Water	0.001	0
Albumin std	0.509	10

Note that the calibration curve is lot dependent.

LIMITATIONS OF THE PROCEDURE

Interference
No known interferences.

MEASURING RANGE
The test has been developed to determine total protein concentrations within a measuring range from 0.5 to 15 g/l.

PERFORMANCE CHARACTERISTICS
The results obtained in individual laboratories may differ from the performance data given. Linearity testing has been performed with water based standard solutions. Different matrixes may change the linearity limits of the test.

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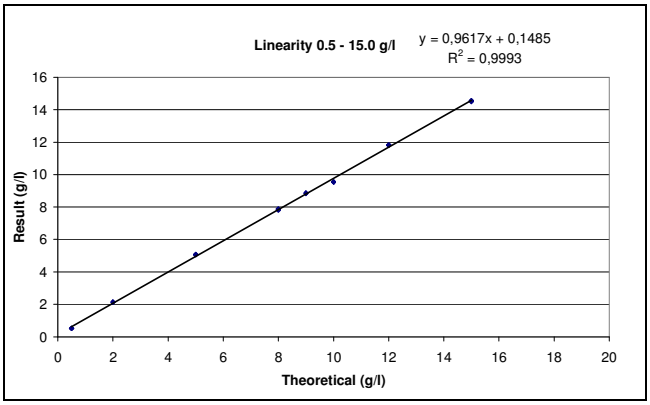

Determination limit (=Test limit low)
The determination limit is the lowest concentration that can be measured quantitatively. The determination limit for this method is 0.5 g/l.

Precision
Gallery analyzer

	Mean 3.5 g/l Dark beer		Mean 4.5 g/l Lager beer		Mean 8.1 g/l Lager beer	
	SD	CV %	SD	CV %	SD	CV %
Within run	0.032	0.9	0.035	0.8	0.062	0.8
Between run	0.076	2.1	0.110	2.4	0.158	1.9
Total	0.083	2.3	0.115	2.6	0.170	2.1

A precision study was performed using Gallery, with the number of measurements being n = 50. Arena analyzer shows similar performance. Samples used for testing were native samples.

Accuracy / Method comparison
Accuracy of the method was tested with spiked native samples. Five spike levels of beer sample were analyzed.

Sample	Result (g/l)	Theoretical value (g/l)	Recovery rate (%)
Beer level 1	3.9	4.0	98
Beer level 2	5.4	5.4	99
Beer level 3	7.3	7.3	100
Beer level 4	8.2	8.3	99
Beer level 5	9.8	9.9	99

OTHER REMARKS
Note that the application performance has been verified with pure chemicals dissolved in deionized water and with spiked native samples. The results obtained in individual laboratories may differ from the given performance data due to e.g. sample matrix, concentrations or analysis environment. Each laboratory is responsible to verify the method to prove the analysis performance.

WASTE MANAGEMENT
Please refer to local legal requirements. It is recommended to empty the analyzer cuvette waste bin and waste water daily. Emptying should be done immediately after the analysis when using hazardous reagents/solutions.
Note: If using reagents/solutions that react with each other, cuvette waste bin and waste water should be emptied and washed between use of these reagents.

BIBLIOGRAPHY
1. Burtis, C.A. und Ashwood, E.R. (Hrsg.), Tietz Fundamentals of Clinical Chemistry, 5th edition, W B Saunders Company, Philadelphia, 2001, S. 326, 1006.
2. Thomas, L. (Hrsg.), Clinical Laboratory Diagnotics; Use and Assessment of Clinical Laboratory Results, 1st edition, TH-Books Verlagsgesellschaft mbH, Frankfurt/Main, Deutschland, 1998, Seiten 644-645.
3. Doumas, B.T., Standards for Total Serum Protein Assays – A Collaborative Study, Clin. Chem. 21/8, 1975, pp. 1159-1166.
4. Guder W.G., Narayanan S, Wisser H, Zawta B. List of Analytes; Preanalytical variables. Brochure in: Samples: From Patient to the Laboratory. GIT Verlag GmbH, Darmstadt, 1996.
5. Young, D.S., Effects of Drugs on Clinical Laboratory Tests, Fifth Edition, AACC Press, Washington, D.C., 2000, pp. 3-672 – 3-677.

ADDITIONAL MATERIAL
Certificate of analysis and SDS are available at
www.e-labeling.eu/TSF

Applications for Gallery and Arena automated analyzers are available upon request from the local sales representative. Information in the Application note can change without prior notice.

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2015-05-20

Changes from previous version
Precautions updated.
General updates.

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