

MICRO PROTEIN

Diagnostic reagent for the in-vitro quantitative determination of micro protein in human cerebrospinal fluid (CSF) and urine on both manual and automated systems.

REF: BS.1/MP02.025.0050	50 test	REF: BS.1/MP02.050.0100	100 test
REF: BS.1/MP02.100.0200	200 test		

CLINICAL SIGNIFICANCE

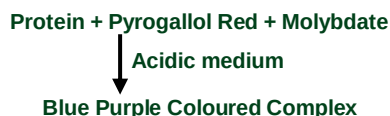
Proteins are involved in the maintenance of the normal distribution of water between blood and the tissues and consist mainly of the albumin and globulin fractions.

Urinary microproteins include microalbumin (mAlb), α 1-microglobulin(A1M), transferrin (TRU), and immunoglobulin G(IgG). These have important diagnostic value in diseases such as diabetic nephropathy, cardiovascular disease caused by rheumatoid arthritis, and hypertensive renal injury.

The presence of protein in urine is a very sensitive indicator of renal disorders. There are four ways by which increased amounts of protein can occur: Increased glomerular permeability, defective tubular reabsorption, increased plasma concentration of an abnormal, low molecular weight protein; and abnormal secretion of protein into the urinary tract.1 Albuminuria, increased amounts of albumin in urine, has been recognized as an early indicator of renal damage in diabetes that can be reversed if detected and treated early. The measurement of CSF total protein provides an indication of either increased permeability of the blood/brain barrier to plasma proteins, or of increased intrathecal secretion of immunoglobulins.

METHOD PRINCIPLE

Proteins, in an acidic medium, combine with Pyrogallol Red and Molybdate to form a blue purple colored complex. Intensity of the color formed is directly proportional to the amount of proteins present in the sample.



REAGENT COMPOSITION

R1: Standard	100 mg/dl
R2: Reagent	
Succinate buffer	100 mmol/L
Sodium oxalate	4.0 mmol/L
Sodium molybdate	60 μ mol/L
Pyrogallol red	80 μ mol/L

PRECAUTIONS AND WARNINGS

Reagent to be handled by entitled and professionally educated person. Do not ingest or inhale as reagents contain sodium hydroxide irritant (xi) chemical and copper (II) sulphate that is an environmentally dangerous reagent.

R36/38: Irritating to eyes and skin.

R52/53: Harmful to aquatic organisms, may cause long-term adverse effects in the aquatic environment

Good Laboratories practices using appropriate precautions should be followed in:

- Wearing personnel protective equipment (overall, gloves, glasses,).
- Do not pipette by mouth.
- In case of contact with eyes or skin; rinse immediately with plenty of soap and water. In case of severe injuries; seek medical advice immediately.
- Respect country requirement for waste disposal.
S56: dispose of this material and its container at hazardous or special waste collection point.
S57: use appropriate container to avoid environmental contamination.
S61: avoid release in environment.

For further information, refer to the micro protein reagent material safety data sheet.

REAGENT PREPARATION, STORAGE AND STABILITY

BioScien Micro protein reagents are supplied ready-to-use. All reagents are stable until expiration date stated on label when properly stored refrigerated at 2-8°C.

Deterioration

The **BioScien** Micro protein reagent is normally clear, do not use reagent if it is turbid.

SPECIMEN COLLECTION AND PRESERVATION

Urine samples collected randomly or 24 hr specimens may be used. Store at 2-8°C or freeze the specimen till assayed.

CSF should be free of hemolysis. Centrifuge any specimen containing red blood cells or particulate matter. CSF may be stored at 2-8°C for several days until assay.

SYSTEM PARAMETERS

Wavelength	600 nm (578 nm)
Optical path	1 cm
Assay type	End-point
Direction	Increase
Sample Reagent Ratio	1:50
e.g.: Reagent volume	1 ml
Sample volume	20 μ l
Temperature	15-25°C
Incubation time	10 min at 15-25°C
Zero adjustment	Reagent blank
Sensitivity	6.0 mg/dL
Linearity	200 mg/dL

EQUIPMENT REQUIRED NOT PROVIDED

- Sterile Syringe
- Analytical tubes, automatic pipet
- Centrifuge and spectrophotometer

ASSAY PROCEDURE

	Blank	Standard	Specimen
Reagent	1.0 ml	1.0 ml	1.0 ml
Standard		20 µl	
Specimen			20 µl

Mix, incubate for 10 minutes at 15-25°C. Measure absorbance of specimen and standard against reagent blank within 10 minutes.

CALCULATION

Micro protein concentration (mg/dl) = $\frac{\text{Abs. specimen}}{\text{Abs. standard}} \times 100$

QUALITY CONTROL

To ensure adequate quality control, it is recommended that normal and abnormal commercial control serum of known concentrations included in each run. If control values are found outside the defined range, check the instrument calibration, and reagent for problems. If control still out of range please contact **BioScien** technical support.

PERFORMANCE CHARACTERISTICS

Precision	Within run (Repeatability)		Run to run (Reproducibility)	
	Normal level	High level	Normal level	High level
n	20	20	20	20
Mean mg/dl	37	105	39	109
SD.	0.74	1.27	0.79	1.36
CV. %	2.0	1.3	2.7	1.9

The results of the performance characteristics depend on the analyzer used.

Accuracy (Methods Comparison)

Result obtained from **BioScien** micro Protein reagent compared with commercial reagent of the same methodology performed on 20 human sera give a correlation of 0.975.

Sensitivity

When run as recommended, the minimum detection limit of the assay is 6.0 mg/dL micro Protein.

Linearity

The reaction is linear up to micro Protein concentration of 200 mg/dl. Specimens showing higher concentration should be diluted 1+1 using physiological saline and repeat the assay (result×2).

INTERFERING SUBSTANCES

- Avoid erythrocyte contamination.
- No significant interference up to uric acid level of 85 mg/dl.
- No significant interference up to oxalate level of 90 mg/dl.
- No significant interference up to phosphate level of 1.2 g/L.
- No significant interference up to calcium level of 130 mg/dl.
- No significant interference up to creatinine level of 6 g/L.
- No significant interference up to ascorbic acid level of 10 mg/dl.
- No significant interference up to bilirubin level of 60 mg/dl.

EXPECTED VALUES ⁽⁵⁾

Sample	
CSF	15-45 mg/dl
Urine (24 hrs.)	20-145 mg/day
Urine (random)	<10 mg/dl

DYNAMIC RANGE

6.0 – 200 mg/dl.

REFERENCES

1. Teitz N.W., Textbook of Clinical Chemistry. W.B. Saunders Co. (1986) pp 602-613. Dilella B.A., Peneberthy L.A, Fraser C.G., Clin Chem. (1983) 29:356-360.
2. Henry R.J., Cannon. D.C., Winkelman J.W., "Clinical Chemistry, principles and Techniques." Harper & Row, 2nd Ed. 1974.
3. Watanabe N., Kamei, S., Oh Kubo A., and Tok uda K., Clin. Chem., 1986.,32/8:1551.

SYMBOLS IN PRODUCT LABELLING			
	For in-vitro diagnostic use		Number of <n> test in the pack
	Batch Code/Lot number		Caution
	Catalogue Number		Do not use if package is damaged
	Temperature Limitation		Consult Instruction for use
	Expiration Date		
	Manufactured by		