Clinical Chemistry

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REAGENT 29 mL

IVD CE

HORIBA ABX SAS
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FRANCE

ABX Pentra Urinary Proteins CP

- **Pentra C400**

Diagnostic reagent for quantitative *in vitro* determination of Total Proteins in urine by colorimetry.

Application Release

Urine: TPU
1.xx

Intended Use

ABX Pentra Urinary Proteins CP reagent is intended for the quantitative *in vitro* diagnostic determination of urinary proteins in urine.
Identification of urinary protein is used in the diagnosis and treatment of disease conditions such as renal or heart diseases or thyroid disorders, which are characterized by proteinuria or albuminuria.

Clinical Interest (1, 2)

Elevated concentration of total protein in urine (proteinuria) can be detected in the majority of kidney diseases. Primary and secondary nephropathies may cause increased glomerular permeability or decreased tubular reabsorption. Post-renal causes of proteinuria are infections, bleedings or malignant diseases of the urinary tract. Elevated urine protein levels can also be related to other acute disorders like fever.

Method

The total protein test for urine is based on the procedure developed by Watanabe *et al.* (3) which is a dye-binding colorimetric method utilizing pyrogallol red-molybdate complex. This photometric test which provides good precision and linearity, has been modified to equalize the reactivity of albumin and gamma-globulin (4).
The pyrogallol red is combined with molybdenum acid, forming a red complex with maximum absorbance at 467 nm. When this complex is combined with protein in acidic conditions, a blue-purple color develops with an increase in absorption at 598 nm (3).
The color is directly proportional to the protein concentration.

Reagents

ABX Pentra Urinary Proteins CP is ready-to-use.

Reagent:	
Pyrogallol red	60 µmol/L
Sodium molybdate	40 µmol/L
Detergents	

ABX Pentra Urinary Proteins CP should be used according to this notice. The manufacturer cannot guarantee its performance if used otherwise.

Handling

1. Remove the cap of the cassette.
2. If present, remove foam by using a plastic pipette.
3. Position the protective cap (GBM0969) on the cassette.
4. Place the cassette into the refrigerated reagent compartment.

Calibrator

For calibration, use:
ABX Pentra TPU Cal (A11A01898) (not included)
3 x 3 mL

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Control [a]

For internal quality control, use:

- **ABX Pentra Urine Control L/H / Yumizen C Urine Level 1 Control** (A11A01674 / 1300023946) (not included)
  1 x 10 mL + 1 x 10 mL / 6 x 5 mL
- **Yumizen C Urine Level 2 Control** (1300023947) (not included)
  6 x 5 mL

Each control should be assayed daily and/or after a calibration.
The frequency of controls and the confidence intervals should correspond to laboratory guidelines and country-specific directives. You should follow federal, state and local guidelines for testing quality control materials. The results must be within the range of the defined confidence limits. Each laboratory should establish a procedure to follow if the results exceed these confidence limits.

Materials Required but not Provided [a]

- Automated clinical chemistry analyzer: Pentra C400
- Calibrator: **ABX Pentra TPU Cal** (A11A01898)
- Controls:
  ABX Pentra Urine Control L/H / Yumizen C Urine Level 1 Control (A11A01674 / 1300023946)
  Yumizen C Urine Level 2 Control (1300023947)
- Standard laboratory equipment.

Specimen

- Urine.

Stability (5): 1 day at 20-25°C
7 days at 4-8°C
1 month at -20°C

Reference Range (6)

Each laboratory should establish its own reference ranges. The values given here are used as guidelines only.

Urine (Excretion):
Adult: < 100 mg/day (< 0.10 g/day)
Pregnancy: < 150 mg/day (< 0.15 g/day)

Storage and Stability

Stability before opening:

Stable up to the expiry date on the label if stored at 2-8°C.

Stability after opening:

Refer to the paragraph "Performance on Pentra C400".

Do not freeze.

Waste Management

Please refer to local legal requirements.

General Precautions [b]

- This reagent is for professional *in vitro* diagnostic use only.
- For prescription use only.
- This reagent is classified as non-hazardous in compliance with regulation (EC) N°.1272/2008.
- **Warning:** This reagent is obtained from substances of animal origin. Consequently, it should be treated as potentially infectious and handled with the appropriate cautions in accordance with good laboratory practices (7).
- Do not pipette by mouth.
- Do not replenish the reagents.
- Do not swallow. Avoid contact with skin and mucous membranes.
- Observe the standard laboratory precautions for use.
- The reagent cassettes are disposable and should be disposed of in accordance with the local legal requirements.
- Please refer to the SDS associated with the reagent.
- Do not use the product if there is visible evidence of biological, chemical or physical deterioration.
- It is the user's responsibility to verify that this document is applicable to the reagent used.

[a]Modification: new control.
[b]Modification: general precautions modification.

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Performance on Pentra C400

Urine

The performance data listed below are representative of performance on HORIBA Medical Systems.
Number of tests: 100 tests

On Board Reagent Stability
Once opened, the reagent cassette placed in the refrigerated Pentra C400 compartment is stable for 30 days.
Sample volume: 5.0 µL/test

Detection Limit
The detection limit is determined according to the Valtec protocol (8) and equals 0.027 g/L (2.7 mg/dL).

Accuracy and Precision

Repeatability (within-run precision)
Repeatability according to the recommendations found in the Valtec protocol (8) with samples tested 20 times:

- 2 controls
- 3 specimens (low / medium / high levels)

	Mean value g/L	Mean value mg/dL	CV %
Control specimen 1	0.19	19	2.50
Control specimen 2	0.37	37	1.24
Specimen 1	0.19	19	2.67
Specimen 2	0.59	59	0.87
Specimen 3	1.16	116	0.44

Reproducibility (total precision)
Reproducibility according to the recommendations found in the CLSI (NCCLS), EP5-A protocol (9) with samples tested in duplicate for 20 days (2 series per day):

- 2 controls
- 2 specimens (low / high levels)

	Mean value g/L	Mean value mg/dL	CV %
Control specimen 1	0.18	18	8.77
Control specimen 2	0.36	36	2.86
Specimen 1	0.24	24	4.25
Specimen 2	1.28	128	2.61

Measuring Range

The assay confirmed a measuring range from 0.03 g/L (3 mg/dL) to 2.70 g/L (270 mg/dL).
The measuring range is extended up to 5.4 g/L (540 mg/dL) with the automatic post-dilution.
The reagent linearity has been assessed up to 2.70 g/L (270 mg/dL) according to the recommendations found in the CLSI (NCCLS), EP6-A protocol (10).

Correlation

Patient samples: urine
Number of patient samples: 115
Specimens are correlated with a commercial reagent taken as reference according to the recommendations found in the CLSI (NCCLS), EP9-A2 protocol (11).
Values ranged from 0.04 g/L (4 mg/dL) to 2.64 g/L (264 mg/dL).
The equation for the allometric line obtained using Passing-Bablok regression procedure (12) is:
Y = 1.05 X - 0.03 (g/L)
Y = 1.05 X - 2.73 (mg/dL)
with a correlation coefficient r^2 = 0.9886.

Interferences[c]

Haemoglobin: Do not use hemolysed samples.
Direct Bilirubin: No significant influence is observed up to 83 µmol/L (4.8 mg/dL).

Other limitations are given by Young as a list of drugs and preanalytical variables known to affect this methodology (13, 14).

Calibration Stability

The reagent is calibrated on Day 0. The calibration stability is checked by testing 2 control specimens.
The calibration stability is 21 days.
Note: A recalibration is recommended when reagent lots change, and when quality control results fall outside the range established.

Conversion Factor:

g/L x 100.0 = mg/dL

Reference

1. Johnson AM, Rohlfs EM, Silverman LM. Proteins. In: Burtis CA, Ashwood ER, editors. Tietz Textbook of Clinical Chemistry. 3rd ed. Philadelphia: WB Saunders Company (1999): 477-540.

[c]Modification: modification of interferences.

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