

Copper (Colorimetric Test with Dibrom-PAESA)

REF: 232 001 (2 x 25 ml) 50 test
REF: 232 002 (4 x 25 ml) 100 test

Intended Use

Spectrum Diagnostics Copper reagent is intended for in-vitro quantitative, diagnostic determination of Copper in human serum, plasma or urine on both manual and automated systems.

Background

Copper (Cu) is an important trace element and is associated with a number of metalloproteins and it is a catalytic component of numerous enzymes and also a structural component of other important proteins. Copper is involved in many vital processes in the body, Energy Production, connective tissue formation, iron metabolism, melanin synthesis, Normal function of CNS, Regulation of gene expression and has Antioxidant function. Excess Cu ingestion interfere with absorption of zinc and can lead to Zinc deficiency, which is frequently characterized by slow healing. The classical presentation of Cu toxicosis is represented by the genetic disease of Cu accumulation known as Wilson's disease. This disease is typified by hepatocellular damage (increase transferase) and/or changes in mood and behavior because of accumulation of Cu in Central Neurons.

Method

Colorimetric with Dibrom-PAESA

Assay Principle

Copper forms with 4-(3,5-dibromo-2-pyridylazo)-N-ethyl-sulfopropylaniline a chelate complex. The increase of absorbance of this complex can be measured and is proportional to the concentration of total copper in the sample.

Reagents

Standard (ST)

100 µg/dL 15.7 µmol/L

R (Monoreagent)

Acetate buffer pH 5.0 0.2 mol/L

4-(3,5-dibromo-2-pyridylazo)-N-ethyl-sulfopropylaniline 0.02 mmol/L

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

Precautions and Warnings

Do not ingest or inhale. 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.

Avoid contamination by using clean laboratory material (pipette, plastic vial for analyzers,...)

Reagent Preparation, Storage and Stability

Warning: The reagent could precipitate during refrigerate storage. It suggested to let it to redissolve at room temperature before use (15 minutes). Mix well after redissolving.

Spectrum Copper reagent is supplied ready-to-use and stable up to the expiry date labeled on the bottles. Once opened, the opened vial is stable for 3 months at 2-8 °C.

SYMBOLS IN PRODUCT LABELLING

	Authorised Representative		Temperature Limitation
	For in-vitro diagnostic use		Use by/Expiration Date
	Batch Code/Lot number		CAUTION. Consult instructions for use
	Catalogue Number		Manufactured by
	Consult instructions for use		

Specimen collection and preparation

Serum, Plasma (free from haemolysis)

24 hours Urine:

Refrigerate or add 10 ml of 3 mol/L HCl to the container before collection.

System Parameters

Wavelength	580 nm (Hg 578)
Optical path	1 cm
Assay type	End-point
Direction	Increase
Temperature	37 °C
Zero adjustment	Reagent blank
Linearity	500 µg/dl (78.65 µmol/l)

Procedure

I- Determination of copper in serum

	Blank	Standard	Sample
Reagent	1.0 ml	1.0 ml	1.0 ml
Standard	50 µl
Sample	50 µl

Mix and incubate for 5 minutes at 37 °C. Measure the absorbance of the sample A_s and of the standard A_{std} against the reagent blank A_{RBL} .

$$\Delta A_s = A_s - A_{RBL}$$

$$\Delta A_{std} = A_{std} - A_{RBL}$$

Calculation

$$\text{Serum Copper conc. (µg/dL)} = \frac{\Delta A_s}{\Delta A_{std}} \times 100$$

$$\text{Serum Copper conc. (µmol/l)} = \frac{\Delta A_s}{\Delta A_{std}} \times 15.7$$

II- Determination of copper in urine

Dilute Standard 20 Times (Example: 50 µl standard + 950 µl normal saline), then follow the method below :

	Blank	Standard	Urine Sample
Reagent	1.0 ml	1.0 ml	1.0 ml
Standard	500 µl
Sample	500 µl
Dist.H2O	500 µl

Mix and incubate for 5 minutes at 37 °C. Measure the absorbance of the sample A_s and of the standard A_{std} against the reagent blank A_{RBL} .

$$\Delta A_s = A_s - A_{RBL}$$

$$\Delta A_{std} = A_{std} - A_{RBL}$$

Calculation

$$\text{Urine Copper conc. } (\mu\text{g/dL}) = \frac{\Delta A_s}{\Delta A_{std}} \times 5$$

$$\text{Urine Copper conc. } (\mu\text{mol/l}) = \frac{\Delta A_s}{\Delta A_{std}} \times 0.785$$

Quality Control

Normal & abnormal commercial control serum of known concentrations should be analyzed with each run.

Linearity

The reaction is linear up to a Copper concentration of 500 $\mu\text{g/dl}$ (78.65 $\mu\text{mol/l}$)

Specimens showing higher concentration should be diluted 1+1 using physiological saline and repeat the assay (result \times 2).

Interfering Substances:

Interferences are found according to the literatures.

Expected Values

In Serum

Adult males	70 - 140 $\mu\text{g/dl}$ (11 - 22 $\mu\text{mol/l}$)
Adult females	80 - 155 $\mu\text{g/dl}$ (12.5 - 24.3 $\mu\text{mol/l}$)
Females in pregnancy	120 - 300 $\mu\text{g/dl}$ (18.8 - 47 $\mu\text{mol/l}$)
Children (6-12 years)	80 - 190 $\mu\text{g/dl}$ (12.5 - 29.8 $\mu\text{mol/l}$)
Infants	20 - 70 $\mu\text{g/dl}$ (3.14 - 11 $\mu\text{mol/l}$)

In 24hours Urine 10 - 30 $\mu\text{g}/24\text{hours}$

Spectrum Diagnostics does not interpret the results of a clinical laboratory procedure; interpretation of the results is considered the responsibility of qualified medical personnel. All indications of clinical significance are supported by literature references.

Waste Disposal

This product is made to be used in professional laboratories. Please consult local regulations for a correct 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. refer to special instructions/safety data sheets.

References

1. Abe A., Yamashita S., Noma A., Clin. Chem., 552-554-35 (1989)
2. Richmond. N., Clin. Chem. 1973; 19: 1350-1356.

ORDERING INFORMATION	
CATALOG NO.	QUANTITY
232 001	50 Test
232 002	100 Test



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