SPECTRUM

The Creative Approach to Bioscience

Copper
(Colorimetric Test with Dibromo-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 absorbtion 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 (increased transferase) and/or changes in mood and behavior because of accumulation of Cu in Central Neurons.

Method

Colorimetric with Dibromo-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 inhalate. 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 refrigeration.It is 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 reagent and the standard vials are stable for 3 months at specified temperature.

Deterioration

Failure to recover control values within assigned range may indicate reagent deteriortion

SYMBOLS IN PRODUCT LABELLING

EC REP Authorised Representative	Use by/Expiration Date
IVD For in-vitro diagnostic use	CAUTION. Consult instructions for use
LOT Batch Code/Lot number	Manufactured by
REF Catalogue Number	(Xi) - Irritant
Consult instructions for use	
Temperature Limitation	

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
Reagent Blank Limits	Low 0.00 AU High 0.17 AU
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 As and of the standard Ast against the reagent blank A_{RBL} .

$\Delta As = As - A_{RBL}$
$\Delta Astd = Astd - A_{RBL}$

Calculation

$$\text{Serum Copper conc. (µg/dL)} = \frac{\Delta As}{\Delta Astd} \times 100$$

$$\text{Serum Copper conc. (µmol/l)} = \frac{\Delta As}{\Delta Astd} \times 15.7$$

II- Determination of copper in urine

Dilute Standard 10 Times (Example: 100 µl standard + 900 µl normal saline),then follow the method below :

	Blank	Standard	Urine Sample
Reagent	.....	1.0 ml	1.0 ml
Diluted Standard	.....	750 µl	.....
Sample	.....	.....	750 µl
Dist.H2O	1.0 ml	.....	.....

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

$\Delta As = As - A_{RBL}$
$\Delta Astd = Astd - A_{RBL}$

Calculation

$$\text{Urine Copper conc. } (\mu g/dL) = \frac{\Delta As}{\Delta Astd} \times 10$$

$$\text{Urine Copper conc. } (\mu mol/l) = \frac{\Delta As}{\Delta Astd} \times 1.57$$

$$\text{Copper conc. } (\mu g/\text{urine 24h}) = \frac{\Delta As}{\Delta Astd} \times 10 \times \text{dl of urine 24h}$$

Quality Control

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

Sensitivity

When run as recommended, the minimum detection limit of this assay is 10.0 µg/dL.

Linearity

The reaction is linear up to a Copper concentration of 500 µg/dl (78.65 µmol/l)
Specimens showing higher concentration should be diluted 1+1 using physiological saline and repeat the assay (result × 2).

Interfering Substances

Interferences are found according to the literatures.

Expected Values

In Serum

Adult males	70 - 140 µg/dl	(11 - 22 µmol/l)
Adult females	80 - 155 µg/dl	(12.5 - 24.3 µmol/l)
Females in pregnancy	120 - 300 µg/dl	(18.8 - 47 µmol/l)
Children (6-12 years)	80 - 190 µg/dl	(12.5 - 29.8 µmol/l)
Infants	20 - 70 µg/dl	(3.14 - 11 µmol/l)

In 24hours Urine **10 - 50 µg/24hours**

Performance characteristics

Accuracy
Results obtained using this reagents (y) did not show systematic differences when compared with other commerical reagents (x).
The results obtained using 92 samples were the following :
correction coefficient (r) : 0.986
Regression equation : y =4.4+0.920x.
The results of the performance characteristics depend on the analyzer used.

Spectrum Diagnostics does not interpret the results of a clinical laboratory procedure; interpretation of the reults 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

Spectrum For Diagnostics Industries - Free Zone
Ismailia Free Zone Industrial Area, Block 5 .
Cairo- Port said Avenue.
Ismailia,Egypt
Tel: +2 064 3488 013 - +2 064 3488 014 Fax: +2 064 3488 015
www.sdi-fz.com

EC REP **MDSS GmbH**
Schiffgraben 41
30175 Hannover, Germany

CE

ISO 13485:2016