



UIBC (Colorimetric Method)

Invitro Diagnostic reagent kit for quantitative determination of Unbound Iron binding Capacity in human serum/plasma sample on Photometric System.



Order Information

Cat. No.	Kit Configuration
OAR1154	Reagent 1: 2 x 40 mL
	Reagent 2: 1 x 12 mL

Summary

The iron is the component of a great number of enzymes. The myoglobin, muscular protein, contains iron, as well as the liver. Iron is necessary for the hemoglobin production, molecule that transports oxygen inside red globules. Their deficit in the last causes the ferropenic anemia. High levels of iron are found in hemochromatosis, cirrhosis, hepatitis and in increased transferrin levels.

Method

Colorimetric Method.

Principle

Serum iron is bound to transferrin, but only about one third of the iron binding sites are saturated with iron. The unsaturated iron-binding capacity of transferrin (UIBC) denotes the available iron-binding sites of serum. The amount of iron that serum transferrin can bind when completely saturated with an excess of Fe⁺³ is the total iron-binding capacity (TIBC).

The method measures the TIBC by first saturating the transferrin with excess of Fe⁺³. The remaining iron is adsorbed with magnesium carbonate, and once the binding process is complete the chelator is removed by centrifugation, and an assay for iron content performed in the supernatant. From this measurement the TIBC value is obtained.

When the serum iron (SI) determination is performed concurrently with the TIBC and the result subtracted from the TIBC value, the difference yields the unsaturated iron-binding capacity (UIBC), or seric transferrin not bound to iron.

Reagents

Storage Instructions and Reagent Stability

Reagent are stable up to the end of the indicated month of expiry, if stored at 2 – 8°C, protected from light and contamination is avoided. Do not freeze the reagents!

Reagent 1: Iron Solution (500 µg/dL)

Reagent 2: Magnesium Hydroxide Carbonate Powder (for 50 Tests)

Composition

Iron Solution 500 µg/dL and Magnesium Hydroxide Carbonate powder.

Warnings and Precautions

- Keep out of reach of children. In case of contact with eyes, rinse immediately with plenty of water and seek

medical advice.

- Take off immediately all contaminated clothing.
- The reagents contain sodium azide (0.95 g/L) as preservative. Do not swallow. Avoid contact with skin and mucous membranes.
- For professional use only!

Waste Management

Please refer to latest Biological Medical Waste (BMW) guidelines and local legal requirements.

Reagent Preparation

All the kit compounds are ready to use and stable until the expiry date stated on the label. Do not use reagents over the expiration date. Store the vials tightly closed, protected from light and prevented contaminations during the use.

Materials required but not provided

NaCl solution 9 g/L

General laboratory equipment

Specimen

Serum, heparin plasma separate at the latest 1h after blood collection from cellular contents.

7 days at 2° –8°C

Assay Procedure

Wavelength 560 nm (540 nm - 580

nm) Optical Path 10 mm

Temperature 37°C

Sample Start

		Ratio = $\frac{\text{Sample}}{\text{R1}} = \frac{1}{2}$
Sample	500 µL	
R1 Buffer Solution	1000 µL	Dilution Factor = 3
<ul style="list-style-type: none"> Mix and allow to stand for 5-20 minutes at room temperature. Add to each tube one scoop (aprox. 100 mg) of R2 and allow to stand for 30 minutes, mixing vigorously at 5-minute intervals. Centrifuge for 10 minutes at 3000 r.p.m. Separate off the clear supernatant. 		
Bring the Iron kit reagents (Cat#ND12100050) to room temperature and proceed with the determination of iron from an aliquot of the supernate as per its IFU.		

Calculation

Total iron-binding capacity (TIBC)

TIBC = µg/dL supernat x 3 (Dilution Factor).

Unbound iron-binding capacity (UIBC) =

TIBC – SI

Transferrin Saturation (%)

$$T_{\text{sat}} = \frac{\text{Serum iron} \times 100}{\text{TIBC}}$$

Conversion Factor

Iron (µg/dL) x 0.179 = Iron (µmol/L)

Quality Controls

For internal quality control any normal and abnormal controls should be assayed with each batch of samples.

Each laboratory should establish corrective action in case of deviations in control recovery.

Performance Characteristics Measuring Range

The test has been developed determine UIBC concentration within a measuring range from 0.66 - 1000 µg/dL. If such value is exceeded the sample should be diluted 1 + 1 with NaCl solution (9 g/L) and results multiplied by 2.

Interferences

No interference was observed by, Ascorbic acid up to 30 mg/dL, Bilirubin up to 40 mg/dL and Triglycerides up to

2000 mg/dL. Contamination of glassware with iron will affect the test. Use acid- washed glassware or plastic tubes.

Sensitivity/Limit of Detection

The lower limit of detection is 0.66 µg/dL.

Linearity

The Linearity of detection is 1000 µg/dL.

Precision

Intra-assay n = 20	Mean (µg/dL)	SD (µg/dL)	C V (%)
Sample 1	107.68	3.14	2.92
Sample 2	185.26	2.32	1.25

Inter-assay n = 20	Mean (µg/dL)	SD (µg/dL)	C V (%)
Sample 1	86.45	3.55	4.11
Sample 2	155.75	1.56	1.00

Method Comparison

A comparison of Nucleus Diagnosys UIBC (y) with a commercially available test (x) using 15 samples gave following results:

$$y = 1.010x - 4.057; r^2 = 0.999.$$

Reference Range

Serum Iron	Women	50 – 170 µg/dL (9.0 - 30.4 µmol/L)
	Men	60 – 175 µg/dL (10.7 - 31.3 µmol/L)
TIBC	Adults	250 - 425 µg/dL (45 - 76 µmol/L)
	Children	100 – 400 µg/dL (18 – 72 µmol/L)
T _{Sat}	Women	15-50%
	Men	20-50%

Each laboratory should check if reference ranges are transferable to its own patient population and determine own reference ranges if necessary.

Literature

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4. Tietz. N.W. Clinical Guide to Laboratory Tests, 3rd Edition. W.B. Saunders Co. Philadelphia, PA. (1995).
5. Central Pollution Control Board (CPCB) Guidelines for Management of healthcare waste as per Biomedical Waste Management rules, 2016. https://www.cpcb.nic.in/uploads/hwmd/Guidelines_healthcare_eJune_2018.pdf

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