IRON (CAB)

INTENDED USE
For the quantitative determination of iron in human serum or heparinized plasma.

CLINICAL SIGNIFICANCE
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. The variation day to day is quite marked in healthy people. Clinical diagnosis should not be made on a single test result; it should integrate clinical and other laboratory data.

METHOD AND PRINCIPLE
Serum iron reacts with chromazurol-B and CTMA-Br to form a complex deeply blue colored. The absorbance measured at 630 nm is directly proportional to the amount of iron in the sample.
The intensity of the color formed is proportional to the iron concentration in the sample.

REAGENT COMPOSITION
R 1 Acetate Buffer pH 4.9 100 mmol/L
R 2 Color CAB 40 mmol/L
IRON CAL Iron aqueous primary standard 200 µg/dL

WARNINGS AND PRECAUTIONS
1. Iron STD: proceed carefully with this product because due its nature it can get contaminated easily.
2. It is recommended to use disposable material. If glassware is used the material should be soaking for 6 h in diluted HCl (20% v/v) and then thoroughly rinsed with distilled water and dried before use.
3. Calibration with the aqueous standard may cause a systematic error in automatic procedures. In these cases, it is recommended to use a serum Calibrator.
4. Use clean disposable pipette tips for its dispensation.
5. The reference values are strongly method dependent.

STORAGE AND STABILITY OF REAGENT.
All the components of the kit are stable until the expiration date on the label when stored tightly closed at 2-8°C, protected from light and contaminations prevented during their use. Do not use reagents over the expiration date.

REAGENT DETERIORATION
Presence of particles and turbidity.
Blank absorbance (A) at 630 nm > 0 .500 abs.

SPECIMEN COLLECTION AND STABILITY
Serum or heparinized plasma. Free of hemolysis and separated from cells as rapidly as possible. Stability of the sample: 2-8°C for 7 days.

INTERFERENCES
Hemolyzed samples are rejected, since erythrocytes contain iron and therefore falsely elevate the serum results. A list of drugs and other interfering substances with iron determination has been reported.

ASSAY PROCEDURE FOR SEMIAUTO ANALYZER.
Wavelength : 630 nm
Temperature :37 deg c

	Blank (µl)	Std (µl)	Test (µl)
Reagent	1000	1000	1000
Std	--	50	--
Test	--	--	50

Mix and incubate the tubes for 10 minutes at 37 deg c.Measure the optical density of standard against reagent blank to generate factor . Multiply test absorbance with the factor to get the IRON results as below.

CALCULATIONS
$$\frac{\text{(A) Sample -- (A) Reagent Blank}}{\text{(A) Standard –(A) Reagent blank}} \times 200\ \mu g/dL$$

QUALITY CONTROL
Use control sera with known normal and abnormal values to monitor the integrity of the reaction. Values should be those acceptable for this method and temperature.

EXPECTED VALUES
Male 65 - 175 µg/dL
Female 40 - 150 µg/dL

PERFORMANCE CHARACTERSTICS
Measuring range: From detection limit of 0.850 µg/dL to linearity limit of 500 µg/dL. If the results obtained were greater than linearity limit, dilute the sample 1/2 with NaCl 9 g/L and multiply the result by 2.

Precision

	Intra-assay (n=20)		Inter-assay (n=20)	
Mean	113	250	111	249
SD	0.89	0.72	3.51	6.29
CV	0.79	0.29	3.17	2.52

Sensitivity: 1ug/dl
Results obtained using competitors reagents did not show systematic differences when compared with other commercial reagents. The results obtained using 50 samples were the following : Correlation coefficient (r)2:0.9934.
Regression equation : y= 1.0243x – 3.877. The results of the performance characteristics depend on the analyzer used**.**

GENERAL TECHNICAL PARAMETERS

Mode	**End point.**
Wavelength	**630 nm**
Reaction direction	**Increasing**
Reagent Blank	**Yes**
Sample Vol	**0.050 ml**
Reagent Vol	**1.000 ml**
Measuring time	**10 min at 37 deg c**
Reagent Blank Abs Max	**NMT 0.500 Abs**
Calibration Mode	**1-point**
Standard Conc	**200 µg/dL**
Linearity	**500 µg/dL**

Decimal Places	**1**
Unit	**µg/dL**
Ref Low	**Male 65 µg/dL /Female 40 µg/dL**
Ref high	**Male 175 µg/dL /Female 150 µg/dL**

REFERENCES

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