Reactivos GPL

Barcelona, España

CE

-Iron FZ-

IRON
Ferrozine. Colorimetric

Conservar entre: +2+8ºC.

Presentation:
Cod. SU022 CONT: R1 4 x 50 mL. + R2 1 x 100 mg. + R3 1 x 10 mL. + Cal 1 x 5 mL

Procedure

Quantitative determination of iron.

Only for *in vitro use in clinical laboratory* (IVD)

TEST SUMMARY
The iron is dissociated from transferring-iron complex in weakly acid medium. Liberated iron is reduced into the bivalent form by means of ascorbic acid. Ferrous ions give with FerroZine a coloured complex:

$$\text{Transferrin } (Fe^{3+})_2 + e^- \xrightarrow{\text{Ascorbic acid}} 2\ Fe^{2+} + \text{Transferrin}$$

$$Fe^{2+} \xrightarrow{\text{FerroZine}} \text{Coloured complex}$$

The intensity of the color formed is proportional to the iron concentration in the sample[1,2].

REAGENTS COMPOSITION

R.1 (Buffer)	Acetate pH 4.9	100 mmol/L
R.2 (Reductant)	Ascorbic acid	99.7%
R.3 (Color)	Ferrozine	40 mmol/L.
Iron Cal	Iron aqueous primary standard 100 µg/dL.	

REAGENT PREPARATION AND STABILITY
- Working reagent (WR): Dissolve (→) the contents of one tube R 2 Reductant in one bottle of R 1 Buffer. Cap and mix gently to dissolve contents.
Stability: 3 months at 2-8ºC or 1 month at 15-25ºC.
All the components of the kit are stable until the expiration date on the label when stored tightly closed at 2-8ºC, protected from light and contamination prevented during their use.

Iron Cal:
Once open is stable up to 1 month when stored tightly closed at 2-8ºC, protected from light and contamination prevented during their use.

Signs of reagent deterioration:
- Presence of particles and turbidity.
- Blank absorbance (A) at 562 nm ≥ 0.020

All the reagents of the kit are stable up to the end of the indicated month and year of expiry. Store tightly closed at 2-*8ºC*. Do not use reagents over the expiration date.

SPECIMEN
Serum or heparinized plasma.
Fee of hemolysis and separated from cells as rapidly as possible.
Stability of the sample: 2-8ºC for 7 days[1].

MATERIAL REQUIRED BUT NOT PROVIDED
- Spectrophotometer or colorimeter measuring at 562 nm.
- Matched cuvettes 1.0 cm. light path.

General laboratory equipment(Note 1).

TEST PROCEDURE
1. Assay Conditions
- Wavelength : ..... 562 nm. (530-590).
- Cuvette: ..... 1 cm light path.
- Temperature .....37ºC. 15-25ºC.

2. Adjust the instrument to zero with distilled water.
3. Pipette into a cuvette:

	WR Blank	Calibrator	Sample Blank	Sample
WR (mL)	1.0	1.0	1.0	1.0
R 3 (drops)	1	1	--	1
Distilled water (µL)	200	--	--	--
Calibrator(Note 2,3) (µL)	--	200	--	--
Sample (µL)	--	--	200	200

4. Mix and incubate 5 min at 37ºC or 10 min at room temperature (15-25ºC).
5. Read the absorbance (A) of calibrator and sample against WR Blank. The colour is stable for at least 30 minutes.

CALCULATIONS

$$\text{Iron } (\mu g/dL.) = \frac{(A)\text{Sample} - (A)\text{Sample Blank}}{(A)\text{Standard}} \times 100 \text{ (Calibrator conc.)}$$

Conversion Factor: µg/dL. x 0.179 = µmol/L.

QUALITY CONTROL
Control sera are recommended to monitor the performance of the procedure, Normal and Pathological.
If control values are found outside the defined range, check the instrument, reagents and calibrator for problems.

Serum controls are recommended for internal quality control. Each laboratory should establish its own Quality Control scheme and corrective actions if controls do not meet the acceptable tolerances.

REFERENCE VALUES[4]

Male	65 - 175 µg/dL ≅ 11,6-31,3 µmol/L(Note 4)
Female	50 - 170 µg/dL ≅ 11,6-31,3 µmol/L(Note 4)

(These values are for orientation purpose).

It is suggested that each laboratory establish its own reference range.

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[1,5,6].
Clinical diagnosis should not be made on a single test result; it should integrate clinical and other laboratory data.

REAGENT PERFORMANCE
- *Measuring Range*:
From detection limit of 5.74 µg/dL. to linearity limit of 1000 µg/dL., under the described assay conditions.
If results obtained were greater than linearity limit, dilute the sample ½ with NaCl 9 g/L. and multiply result by 2.

- *Precision*:

	Intra-assay n= 20		Inter-assay n= 20	
Mean (µg/dL)	103	190	107	192
SD	3.02	1.31	1.38	1.64
CV (%)	2.91	0.69	1.29	0.85

- Sensitivity:
1 µg/dL. = 0.0002 A
- Accuracy:
Results obtained GPL reagents did not show systematic differences when compared with other commercial reagents.
The results of the performance characteristics depend on the analyzer used.

INTERFERING SUBSTANCES
- Hemolyzed samples are rejected, since erythrocytes contain iron and therefore falsely elevate the serum results[1,2].
- A list of drugs and other interfering substances with iron determination has been reported by Young et. al[3,4].

NOTES
1. 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.
2. Calibration with the aqueous standard may cause a systematic error in automatic procedures. In these cases, it is recommended to use a serum Calibrator.
3. Use clean disposable pipette tips for its dispensation.
4. Strongly method dependent.

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