IRON – FERROZINE IVD CE

PRODUCT CODE
CS028

INTENDED USE
Reagents for measurement of iron concentration Only for in vitro use in the clinical laboratory

CLINICAL SIGNIFICANCE
Following intestinal absorption of iron or erythrocyte destruction, iron ions are released into the plasma where they bind to either apotransferrin or apoferritin proteins to form transferrin and ferritin, respectively. The former helps transport iron to bone marrow for erythropoiesis; the latter stores iron in tissues, until is needed. An increase in the iron level in plasma due to rapid destruction of erythrocytes or excesive uptake of iron may also lead to iron overload. The latter causes iron deposition disorders in tissue known as hemosiderosis or hemochromatosis. Conversely, a decrease in the iron level in plasma due to malnutrition or malapsorption may lead to excesive depletion in iron storage, resulting in anemia such as iron-deficiency anemia.

PRINCIPLE
The Fe+3 bound to serum ferritine once dissociated in a week-acid medium by Teepol and guanidium chloride, is reduced by hydroxylamine to Fe+2, forming the ferrous ion a colored complex with FerroZine® proportional to the concentration of iron present in the sample.

$$\boldsymbol{Transferrin} - Fe^{2} \xrightarrow[\textbf{Teepol}]{PH < 5} Apotransferrin$$

$$\boldsymbol{Fe^{3} + Hydroxylamine} \longrightarrow \boldsymbol{Fe^{2}}$$

$$\boldsymbol{Fe^{2} + FerroZine} \longrightarrow \textbf{Fe (FerroZine)}_{3}^{2}\ \textbf{complex}$$

REAGENT COMPOSITION

Iron REAGENT 1 (Buffer)

Guanidine chloride	1.0 mol/L
hydroxylamine	0.6 mol/L
acetate buffer	400mmol/L
pH	4.0

Iron REAGENT 2

(Chromgen)	
FerroZine	8 mmol/L
Sodium acetate	400 mmol/L

Iron STANDARD

Ferric ion	100 $\mu g/dl$ (17.9 $\mu mol/L$)

REAGENT PREPARATION
Working reagent. Mix 4 volumes of R1 + 1 volume of R2. Stable 6 months at 2-8ºC.

REAGENT STORAGE AND STABILITY
Store at 2-8ºC. All the kit compounds are 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. Discard If appear signs of deterioration: - Presence of particles and turbidity. - Blank absorbance (A) at 560 nm > 0.050 in 1cm cuvette.

SPECIMEN
Serum or heparinized plasma. Centrifuge specimen as soon as possible after collection. Hemolyzed samples are rejected. Ruptured red cells falsely elevate the serum results. Iron in serum is stable for 3 weeks at 2-8ºC and for about 7 days at 20-25ºC. Freeze for longer storage.

PRECAUTION
To avoid contamination, use clean laboratory wares.
Serum specimens should be considered infectious and handled appropriately.

PROCEDURE
1. Bring the Reagent to room temperature.
2. Pipette into cuvettes

Tubes	Reagent Blank	Sample blank	Sample	CAL Standard
Distilled water	200 µL			
Standard				200 µL
Sample	--	200 µL	200 µL	-
Reagent(1)	--	1000 µL	--	--
working reagent	1000 µL		1000 µL	1000 µL

3. Mix and let the tubes stand 5 minutes at room temperature.
4. Read the absorbance (A) of the sample blank at 560 nm against distilled water.
5. Read the absorbance (A) of the samples and the standard at 560 nm against the reagent blank.

CALCULATION
The iron concentration in the sample is calculated using the following general formula:

$$\frac{\text{A Sample} - \text{A Sample blank}}{\text{A Standard}}\ x\ C\ Standard = \mu g/dl\ iron$$

Samples with concentrations higher than 1000 µg/dL should be diluted 1:2 with saline and assayed again. Multiply the results by 2.
If results are to be expressed as SI units apply: µg/dL x 0.179 = µmol/L

LINEARITY
From detection limit of 2.69 $\mu g/dl$ to linearity limit up to 1000 $\mu g/dl$
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.

Interferences:
-Lipemia (intralipid >1.25 g/L) may affect the results.
-Bilirubin (< 40 mg/dL) does not interfere.
-Hemoglobin may affect the results.
- Other drugs and substances may interfere.

NORMAL RANGE
Serum
Men: 65 - 175 µg/dL = 10.7 – 31.3 µmol/L

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Women: 50 - 170 µg/dL = 9.0 – 30.4 µmol/L

QUALITY CONTROL
All control sera with IRON value estimated by this method can be used. Each laboratory should establish its own Quality Control scheme and corrective actions if controls do not meet the acceptable tolerances.

NOTES
-Contamination of glassware with iron will affect the test. Use acid-washed glassware or plastic tubes.
-This method may be used with different instruments. Any application to an instrument should be validated to demonstrate that results meet the performance characteristics of the method. It is recommended to validate periodically the instrument. Contact to the distributor for any question on the application method.
- Clinical diagnosis should not be made on findings of a single test result, but should integrate both clinical and laboratory data.

SYMBOL ON LABELS

BIBILOGRAPHY
1. Carter, P. Anal. Biochem. 40 : 450 (1971).
2. Artiss, J.D., Vinogrador, S., and Zak, B. Clin. Biochem. 14 : 311 (1981).
3.Young DS. Effects of drugs on clinical laboratory tests, 5th ed. AACC Press, 2000.
4. Tietz, N.W., Fundamentals of Clinical Chemistry, p.940. W.B. Saunders Co., Philadelphia , 1987.