ISO 13485:2016 ISO 9001:2015

Ver.250101

Tissue Iron Content Assay Kit

BC 4350 (50 Tests/48 Samples)

FOR RESEARCH USE ONLY, DO NOT USE IT IN CLINICAL DIAGNOSIS

Principle

Iron, bound to Transferrin in Fe(III) form, is released in an acidic medium and the Ferric ions are reduced to Ferrous ions. The Fe (II) ions react with Cromazurol B to form an intensely coloured complex. Intensity of the colour formed is directly proportional to the amount of Iron present in the sample. Wavelength measured at 630 nm

Kit components

Reagent / Component	Volume	Storage
Extraction Reagent	1 × 60mL	2-8°C
Reagent 1	2 × 50 mL	2-8°C
Standard	1 × 4 mL	2-8°C

Unit Conversion

Traditional Unit	SI Unit	Conversion from Traditional to SI
µg/dL	µmol/L	× 0.1791

Reagent Preparation

Iron Reagent and Iron Standard are ready to use

Reagent Storage and Stability

The sealed reagents are stable up to the expiry date stated on the label, when stored at 2-8°C and protected from light.

Open Vial Stability

Once opened, the reagent is stable up to 4 weeks, if contamination is avoided.

Reagent Deterioration

Turbidity or precipitation in any kit component indicates deterioration and the component must be discarded. Values outside the recommended acceptable range for the control may also be an indication of reagent instability and associated results are invalid. Sample should be retested, using a fresh vial of reagent.

Precaution

- To avoid contamination, use clean laboratory wares. Use clean, dry disposable pipette tips for dispensing. Close reagent bottles immediately after use.
- Avoid direct exposure of reagent to light.
- Do not blow into the reagent bottles.
- This reagent is only for IVD use and follow the normal precautions required for handling all laboratory reagents.

Sample Preparation

1. Bacteria or cells

Harvest the cells and wash twice with PBS. Ideal to use 5 million cells for the assay, Add 1mL. Extraction Reagent to 5 million cells and ultrasonicate (200W, work time 3 second/interval 10 second repeat for 30 times) for complete lysis. Perform ultrasonication while keeping the cells in ice bath. Centrifuge at 8000 rpm, 4°C for 10 minutes and collect the supernatant. The supernatant should be kept on ice.

Note: Ideal proportion of Cells/Bacteria to Extraction Reagent is 1:5-10.

2. Tissue

Prepare 10% tissue homogenase by adding 1 mL Extraction Reagent to 0.1g tissue Grind completely to make a homogenate. Centrifuge at 5000 rpm, 4°C for 10 minutes and collect the supernatant

3. Serum or Plasma

Directly use for the assay.

Materials Required but Not Provided

- Pipettes & Tips
- Test Tubes & racks
- Timer
- Incubator
- Analyzer

Procedure notes

Laboratory Procedure for Semi Auto Analyzer			
	Blank	Standard	Sample
Reagent	1000 µL	1000 µL	1000 µL
Distilled water	40 µL	-	-
Standard	-	40 µL	-
Sample	-	-	40 µL
Mix and incubate for 10 minutes at room temperature. Measure the absorbance (A) of standard and sample against the reagent blank at 630 nm. The colour is stable for 1 hour when protected from light.			

Calculation

$$\text{Iron (µg/dL)} = \frac{\text{Absorbance of Sample}}{\text{Absorbance of Standard}} \times 200$$