(FOR RESEARCH USE ONLY. DO NOT USE IT IN CLINICAL DIAGNOSIS!)

Iron Assay Kit

Catalog No.: BC00076

Size: 100T

Please read the instructions carefully before use. If you have any questions or need further help during experiment, please don't hesitate to contact us through the following methods:

✉ Email (Sale) order@enkilife.com

✉ Email (Techsupport) techsupport@enkilife.com

☎Tel: 0086-27-87002838

Website: www.enkilife.com

Shelf life: Please refer to the label on the outer package.

Techsupport: In order to provide you with better service, please inform us the lot number on the label of the outer package.

Basic Information

Product Name	Iron Assay Kit
Detection Method	Colorimetric
Sample Type	Serum, plasma, tissue
Assay Type	Quantitative
Detection Instrument	Microplate reader (560 nm)
Range	3.125-50μM
Sensitivity	0.8μM

Product Introduction

Iron is one of the essential trace elements for the human body and is crucial for physiological processes such as oxygen transport and fat oxidation. The determination of iron ion content is of great significance for evaluating nutritional status, diagnosing iron metabolism-related diseases, and studying the role of iron in biological processes.

Product Features

• This kit is convenient and quick. The absorbance measurement can be carried out 3-5 minutes after adding the sample to be tested. Usually 10-20 samples can be tested in more than ten minutes.

Principle

Based on the coordination effect between tetrasodium dimethylol orange and Fe^{3+}. When tetrasodium dimethylol orange is added to a solution containing Fe^{3+}, it can form a stable orange-red complex with Fe^{3+}. It can be used to quantitatively detect the concentration of trivalent iron ions. In the absence of Fe^{3+}, tetrasodium dimethylol orange is usually yellow or orange; in the presence of Fe^{3+}, the color of the solution will turn to red. There is an absorption peak at 560nm, and the content of Fe^{3+} can be calculated by measuring the absorbance at this wavelength.

$$Fe^{3+} + 3XO \rightarrow Fe(XO)_3$$

Components

No.	Components	Size (100T)	Storage
Reagent 1	Detection Buffer	20 mL	-20°C, protected from light, store at 2-8°C after opening.
Reagent 2	Color Development Solution	15 mL	-20°C, protected from light, store at 2-8°C after opening.
Reagent 3	10 mmol/L Iron Ion Standard	2 mL	-20°C, protected from light, store at 2-8°C after opening.
Consumable 1	Microplate	1 plate	RT
Consumable 2	Plate Sealer	2 pieces	RT

Storage

The unopened kit can be stored at -20℃ for 12 months. After opening, it can be stored at 2-8 ℃ for 6 months.

Preparation

- **Sample handling**

1. Liquid samples such as serum and plasma: If the serum sample is turbid, it can be diluted with detection buffer before use.
2. Tissue sample: Take 0.1g of fresh tissue block, add 1ml of detection buffer to homogenize, centrifuge at 12000 x g for 10min, and take the supernatant for later use.

Note: Do not use iron utensils to handle or transfer samples.

- **Preparation of the kit**

1. Equilibrate the reagents in the kit to room temperature.
2. Dilution of different concentrations of standards: First, dilute the 10mmol/L iron ion standard to 100 μmol/L with the detection buffer, and then dilute it to different concentrations such as 50, 25, 12.5, 6.25, 3.125, and 0 (blank well) μmol/L using the

detection buffer according to the half-dilution method.

Operation process

1. Standard wells: Take 50 μL of different concentrations of standards and add them to the corresponding wells of the microplate; Measurement wells: Take 50μL of the sample to be tested and add it to the corresponding wells of the microplate.
2. Add 150 μL of color development solution to each well in step (1) and mix by gently pipetting.
3. Measure the OD value at 560 nm using a microplate reader.

Note: When adding reagents to the ELISA wells, they should be added to touch the bottom of the ELISA plate; add samples slowly to avoid bubbles (bubbles will affect the measurement results).

The operation table is as follows:

	Standard tube (well)	**Measurement tube (well)**
Different concentrations of iron ion standards (μL)	50	--
Sample to be tested (μL)	--	50
Color development solution (μL)	150	150
Mix by gently pipetting and oscillating, and measure the OD value at 560 nm using a microplate reader.		

Calculation

Standard fitting curve: y = ax + b

The formula for calculating the iron ion concentration in the sample is:

Fe^{3+}Content (μmol/L) = (ΔA_{560} - b) ÷ a × f

y: standard OD value - blank OD value

x: concentration of the standard

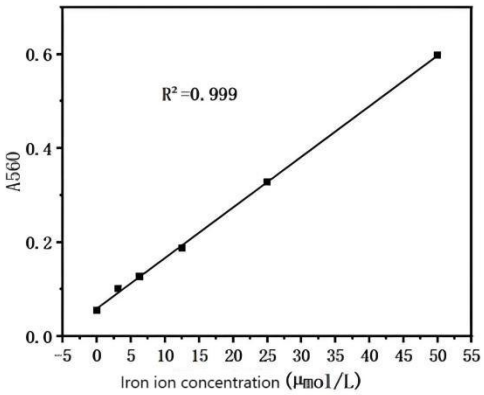
a: slope of the curve

b: intercept of the curve

ΔA_{560}: Sample OD value - Blank OD value

f: dilution factor of the sample before adding it to the detection system

The following standard c



Notes

1. Reagents that are red or nearly red under acidic conditions will interfere with the detection of this kit and should be avoided as much as possible.
2. Substances that affect redox reactions, such as DTT and mercaptoethanol, should not be added to the sample, and detergents such as Tween, Triton and NP-40 should not be added.
3. The measurement requires an ELISA reader capable of measuring A560 or a spectrophotometer capable of measuring trace samples.
4. This product is intended for scientific research use only by professionals and must not be used for clinical diagnosis or treatment, in food or drugs, or stored in ordinary residences.