COD 31934 1 x 15 mL	COD 31935 1 x 45 mL
STORE AT 2-8ºC	
Reagents for measurement of ferritin concentration Only for *in vitro* use in the clinical laboratory	

FERRITIN

CE

BioSystems
REAGENTS & INSTRUMENTS

FERRITIN
LATEX

PRINCIPLE OF THE METHOD

Serum ferritin causes agglutination of latex particles coated with anti-human ferritin antibodies. The agglutination of the latex particles is proportional to the ferritin concentration and can be measured by turbidimetry[1].

CONTENTS

	COD 31934	COD 31935
A. Reagent	1 x 10 mL	1 x 30 mL
B. Reagent	1 x 5 mL	1 x 15 mL

COMPOSITION

A. Reagent: Glycine buffer 170 mmol/L, sodium chloride 100 mmol/L, sodium azide 0.95 g/L, pH 8.2.

B. Reagent: Suspension of latex particles coated with anti-human ferritin antibodies, sodium azide 0.95 g/L.

STORAGE

Store at 2-8ºC.

Reagents are stable until the expiry date shown on the label when stored tightly closed and if contaminations are prevented during their use.

Indications of deterioration:

– Reagents: absorbance of the blank over 1.600 at 540 nm.

AUXILIARY REAGENTS

S. Ferritin Standard: 1 x 3 mL (BioSystems Cod. 31127). Human serum. Ferritin concentration is given on the label. The concentration value is traceable to the Biological Reference Material WHO 94/572 (National Institute for Biological Standards and Control, NIBSC).

Human serum used in the preparation of the standard has been tested and found to be negative for the presence of antibodies anti-HIV and anti-HCV, as well as for HBs antigen. However, the standard should be handled cautiously as potentially infectious.

Reconstitute with 3.00 mL of distilled water. Stable for 1 month at 2-8ºC.

Calibration curve: Prepare dilutions of the Ferritin Standard using 9 g/L saline as diluent. Multiply the concentration of the Ferritin Standard by the corresponding factor indicated below to obtain the ferritin concentration of the dilutions (Note 2).

DILUTION	1	2	3	4	5
Ferritin Standard (µL)	30	60	120	180	240
Saline (µL)	210	180	120	60	–
Factor	0.125	0.25	0.5	0.75	1.0

REAGENT PREPARATION

Working Reagent: Pour the contents of a Reagent B vial (Note 1), into a Reagent A bottle. Mix thoroughly. Stable for 30 days at 2-8ºC. Smaller Working Reagent volumes can be prepared by mixing: 1 mL of Reagent B + 2 mL of Reagent A. Shake the Reagent B vial before pipetting.

ADDITIONAL EQUIPMENT

– Thermostatic water bath at 37ºC

– Analyzer, spectrophotometer or photometer able to read at 540 ± 20 nm.

SAMPLES

Serum collected by standard procedures. Hemolyzed or lipemic samples are not suitable for testing.

Ferritin in serum is stable for 7 days at 2-8ºC.

PROCEDURE

1. Bring the Working Reagent and the instrument to 37ºC.
2. Zero the instrument with distilled water (Note 3)
3. Pipette into a cuvette:

Working Reagent	1.0 mL
Standard (S) or Sample	30 µL

4. Mix and insert cuvette into the instrument. Start stopwatch.
5. Record the absorbance at 540 nm after 10 seconds (A_1) and after 5 minutes (A_2).

CALIBRATION

Calibration curve: Calculate the absorbance difference ($A_{Standard} - A_{Blank}$) of each point of the calibration curve and plot the values found against the ferritin concentration. Ferritin concentration in the sample is calculated by interpolation of its absorbance ($A_{Sample} - A_{Blank}$) on the calibration curve (see Note 2).

A calibration is recommended at least every 7 days, after reagent lot change or as required by quality control procedures.

REFERENCE VALUES

Serum[2,3]

Children: 7-140 µg/L
Men: 20 - 250 µg/L
Women: 20 - 200 µg/L

This range is given for orientation only; each laboratory should establish its own reference range.

QUALITY CONTROL

It is recommended to use the Protein Control Serum levels I (cod. 31211) and II (cod. 31212) to verify the performance of the measurement procedure.

Each laboratory should establish its own internal Quality Control scheme and procedures for corrective action if controls do not recover within the acceptable tolerances.

METROLOGICAL CHARACTERISTICS

– Detection limit: 4 µg/L ferritin

– Measurement interval: (approximate depending on the standard concentration): 4-500 µg/L. For higher values dilute sample 1/5 with 9 g/L NaCl and repeat measurement. Linearity may considerably vary depending on the instrument used (Note 2).

– Repeatibility (within run):

Mean concentration	CV	n
61 µg/L	2.2 %	20
145 µg/L	1.6 %	20

– Reproducibility (run to run):

Mean concentration	CV	n
61 µg/L	3.7 %	25
145 µg/L	1.6 %	25

– Trueness: Results obtained with this reagent did not show systematic differences when compared with reference reagents. Details of the comparison experiments are available on request.

– Zone effect: > 30,000 µg/L.

– Interferences: Hemoglobin (10 g/L), lipemia (triglycerides 5 g/L), bilirubin (62 mg/dL) and rheumatoid factors (520 IU/mL) do not interfere. Other drugs and substances may interfere[4].

These metrological characteristics have been obtained using an analyzer. Results may vary if a different instrument or a manual procedure are used.

DIAGNOSTIC CHARACTERISTICS

Ferritin is the major iron storage compound in the body. It consists of a protein shell enclosing a core of a variable amount of iron. Ferritin is present at particularly high concentrations in liver, bone marrow and spleen.

The plasma ferritin is in equilibrium with body stores and variations in the quantity of iron in the storage compartment are reflected in plasma ferritin concentration.

Serum ferritin concentration declines very early in the development of iron deficiency and it serves as a very sensitive indicator of iron deficiency. On the other hand, a large number of chronic infections, chronic inflamatory disorders (rheumatoid arthritis, renal disease) and malignancies (lymphomas, leukemias, breast cancer, neuroblastoma) result in increased serum ferritin concentration. Plasma ferritin is also increased in patients with hemosiderosis or hemochromatosis[3,5,6].

Clinical diagnosis should not be made on the findings of a single test result, but should integrate both clinical and laboratory data.

NOTES

1. Shake the Reagent B vial gently before using.
2. The calibration curve is linear up to 300 µg/L in some instruments. In these cases, calibration may be performed with a single point (aprox.125 µg/L).
3. These reagents may be used in several automatic analysers. Instructions for many of them are available on request.

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