

COD 31322 1 x 20 mL	COD 31922 1 x 50 mL	COD 31030 2 x 200 mL
STORE AT 2-8°C		
Reagents for measurement of RF concentration Only for <i>in vitro</i> use in the clinical laboratory		

RHEUMATOID FACTORS (RF)



RHEUMATOID FACTORS (RF) LATEX

PRINCIPLE OF THE METHOD

Rheumatoid factors (RF) cause agglutination of the latex particles coated with human gamma-globulin. The agglutination of the latex particles is proportional to the RF concentration and can be measured by turbidimetry¹⁻³.

CONTENTS

	COD 31322	COD 31922	COD 31030
A. Reagent	1 x 16 mL	1 x 40 mL	2 x 160 mL
B. Reagent	1 x 4 mL	1 x 10 mL	2 x 40 mL

COMPOSITION

A. Reagent: Tris buffer 20 mmol/L, sodium azide 0.95 g/L, pH 8.2.

B. Reagent: Suspension of latex particles coated with human gamma-globulin, sodium azide 0.95 g/L.

STORAGE

Store at 2-8°C.

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: Presence of particulate material, turbidity, absorbance of the blank over 1.400 at 650 nm.

AUXILIARY REAGENTS

S. RF Standard: 1 x 3 mL (BioSystems Cod. 31116). Human serum. RF concentration is stated on the vial label. The concentration value is traceable to the WHO Reference Material W1066 (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 3.0 mL of distilled water. Stable for 1 month at 2-8°C.

Calibration curve: Prepare dilutions of the RF Standard using 9 g/L saline as diluent. Multiply the concentration of the RF Standard by the corresponding factor indicated below to obtain the RF concentration of the dilutions (Note 1).

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

ADDITIONAL EQUIPMENT

– Thermostatic water bath at 37°C

– Analyzer, spectrophotometer or photometer thermostatable at 37°C able to read at 650 ± 20 nm.

SAMPLES

Serum collected by standard procedures.

RF in serum is stable for 7 days at 2-8°C.

PROCEDURE

1. Bring the Reagents and the instrument to 37°C.
2. Zero the instrument with distilled water (Note 2).
3. Pipette into a cuvette:

Reagent A	0.8 mL
Water (Blank), Standard (S) or Sample	10 μL
Reagent B (Note 3)	0.2 mL

4. Mix and insert cuvette into the instrument. Start stopwatch.
5. Read the absorbance at 650 nm after 2 minutes of the Reagent B addition.

CALIBRATION

Calibration curve: Calculate the absorbance difference ($A_{\text{Standard}} - A_{\text{Blank}}$) of each point of the calibration curve and plot the values found against the RF concentration. Rheumatoid factors concentration in the sample is calculated by interpolation of its absorbance ($A_{\text{Sample}} - A_{\text{Blank}}$) on the calibration curve.

A calibration is recommended at least every 2 months, after reagent lot change or as required by quality control procedures.

REFERENCE VALUES

Serum, adults⁴: Up to 30 IU/mL.

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

QUALITY CONTROL

It is recommended to use the Rheumatoid Control Serum level I (Cod. 31213) and II (Cod. 31214) 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: 2 IU/mL.

– Measurement interval: (approximate value dependent on the highest standard concentration): 2-160 IU/mL. For higher values dilute sample 1/5 with distilled water and repeat measurement (Note 4).

– Repeatability (within run):

Mean concentration	CV	n
24 IU/mL	5.3 %	20
39 IU/mL	5.6 %	20

– Reproducibility (run to run):

Mean concentration	CV	n
24 IU/mL	6.6 %	25
39 IU/mL	6.1 %	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: This method has not zone effect up to 800 IU/mL.

– Interferences: Hemoglobin (10 g/L), bilirubin (20 mg/dL) and lipemia (triglycerides 10 g/L) do not interfere. Other drugs and substances may interfere⁵.

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

DIAGNOSTIC CHARACTERISTICS

Rheumatoid Factors (RF) are a group of IgM antibodies (although IgG and IgA have been also described) directed against the Fc fragment of the IgG molecules.

RF is mainly present in the serum of patients with rheumatoid arthritis but other diseases may also produce RF: chronic inflammatory processes, infectious diseases such as subacute bacterial endocarditis, malaria, syphilis, leprosy, leishmaniasis, tuberculosis and a variety of autoimmune diseases such as systemic lupus erythematosus⁶.

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

NOTES

1. The calibration curve is linear up to 120 IU/mL in some instruments. In these cases, calibration may be performed with a single point (40 IU/mL). If better accuracy is required, it is recommended to use the multipoint calibration method.
2. These reagents may be used in several automatic analysers. Instructions for many of them are available on request.
3. Shake the Reagent B vial gently before using.
4. The measurement interval depends on the sample to reagent ratio. The interval will be higher by decreasing the sample volume, although the sensitivity of the test will be proportionally decreased.

BIBLIOGRAPHY

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