RF TURBILATEX

(Latex-turbidimetry)

Quantitative Determination of Rheumatoid Factors(RF)

PRINCIPLE OF THE METHOD

The RF-Turbilatex is a quantitative turbidimetric test for the measurement of Rheumatoid factors (RF) in human serum or plasma.

Latex particles coated with human gamma globulin are agglutinated when mixed with samples containing RF. The agglutination causes an absorbance change, dependent upon the RF contents of sample that can be quantified by comparison from a calibrator of known RF concentration.

CLINICALSIGNIFICANCE

Rheumatoid factors are a group of antibodies directed to determinants in the Fc portion of the immunoglobulin G molecule .Although rheumatoid factors are found in a number of rheumatoid disorders, such as systemic lupus erythematosus (SLE) a and Siogren'ssyndrome as well as non-rheumatic condition, its central role in clinic lies its utility as an aid in the diagnosis of rheumatoid arthritis (RA).

REAGENT PROVIDED

Diluent (R1) **:-** 2x 20 ml
Latex (R2) **:-** 2 x 5 ml
RF Calibrator **:-** 1 x 0.5 ml (Ready to use)

WORKING REAGENT PREPARATION

Swirl the latex vial gently before use. Prepare the necessary amount as follows:

0.4 ml of activation Buffer (R1) + 0.1 ml of Latex Reagent (R2)

STORAGE AND STABILITY

All the components of the kit are stable until the expiration date on the label when stored at 2-8°C and contaminations are prevented during their use. Do not use reagents over the expiration date.**Do not freeze the reagents.**

REAGENT DETERIORATION:

Presence of particles and turbidity.

SAMPLES

Fresh serum or plasma. Stable for 7 days at 2-8°C or 3 months at -20°C. The samples with presence of fibrin should be centrifuged before testing. Do not use hemolized or lipemic samples.

CALIBRATION

Use RF Liquid Stable Calibrator provided in the kit.

CALIBRATION CURVE

Single Point Calibration Curve

Dilute the RF Calibrator in 1:1 with NS to obtain new concentration. Calibrate the analyzer using this as calibrator using descriv\bed methid procedure.

Multipoint Calibration Curve

Prepare the following RF Calibrator conc. In 0.9% saline by serial dilution as per the comc. stated on the calibration vial.

StdCalibrator Dilution	5	4	3	2	1
0.9% Saline (µl)	-	200	200	200	200
RF Calibrator (µl)	200	200	200	200	200
Total Volume (µl)	500	200	200	200	400
Diluted Calibrator Curve Conc.	150	75	37.5	18.75	9.375

Note: Prepare the dilution as per the volume required and conc. Stated on the vial label.

PRECAUTIONS

Components from human origin have been tested and found to be negative for the presence of HBsAg, HCV and HIV (1/2). However, handle cautiously as potentially infectious.

SYSTEM PARAMETER

Reaction type	End Point
Wavelength	660 nm
Flow cell Temperature	37°C
Reaction Type	Increasing
Reagent Volume	1000 µl (800+200)
Calibrator/ Sample Vol	15 µl

Incubation Time	300secs (outside at 37°C)
Read Time	5secs
Reagent Blank Abs.	< 1.0Abs
Linearity	Up to 155 IU/mL

PROCEDURE

Manual Method (For Semi-Automatic Analyzers)

1. Allow the reagents and samples to reach room temperature before use.
2. Assay Condition
   Wavelength: 660 nm
   Temperature: 37°C
   Cuvette Light Path: 1 cm
3. Adjust the instrument to zero with distilled water.
4. Pipette into cuvette:

	Blank	Calibrator	Sample
Reagent 1	800 µl	800 µl	800 µl
Reagent 2	200 µl	200 µl	200 µl
D. Water	15 µl	-	-
Calibrator	-	15 µl	-
Serum/Plasma	-	-	15 µl

Mix well and incubate for 5 min at 37°C. Read the absorbance (A) after incubation immediately. Sample & standard incubation time should be same (different incubation times give different absorbance values).

Method (For Fully Automatic Analyzers)

Assay Condition
Wavelength: 660 nm
Temperature: 37°C
Read against water balnk:

Reagent 1	240 µl
Sample	4.5 µl
Mix and after 4.70 minutes of incubation and R2	
Reagent 2	60 µl
Mix and read absorbance (A1) at 6 secs and second absorbance (A2) at 236 seconds.	

CALCULATIONS

$$\frac{(A_2 - A_1)\ \text{sample}}{(A_2 - A_1)\ \text{calibrator}} \times \text{calibrator concentration} = \text{IU/mL RF}$$

REFERENCE VALUES

Normal values up to 30 IU/mL.
Each laboratory should establish its own reference range.

PERFORMANCE CHARACTERISTICS

1. **Linearity Limit:** 0to 155IU /mL, under the described assay conditions. Samples with higher concentrations, should be diluted 1/6 in 0.9% saline and retested again.
2. **Detection Limit:** Values less than 1.5 IU/mL give non-reproducible results.
3. **Prozone Effect:** No prozone effect was detected up to 800 IU/mL.
4. **Sensitivity:** Δ3.34mA.IU/mL.

QULATITY CONTROL

Control Sera (Level L and Level H) are recommended to monitor the performance of manual and automated assay procedures.

INTERFERENCES

Bilirubin (20 mg/dl), Lipemia (10 g/l) and Rheumatoid Factors (300 IU/ml), do not interfere. Hemoglobin (≥ 5 g/l), interferes. Other substances may interfere.

KEE DIAGNOSTICS PVT LTD.
CIN: U24231DL2004PTC128343 .
A Subsidiary of KEE PHARMA LTD.
A-8, 3RD FLOOR NARAINA INDUSTRIAL AREA,PHASE-II,
NEW DELHI-110028
E:-info@keediagnostics.com
T: - 01143136000; W:- www.keediagnostics.com