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Rheumatoid Factor (RF) Latex Test

INTENDED USE

The PULSE RF Latex Test (RF TEST) is intended to be used for the qualitative screening and semi-quantitative determination of Rheumatoid Factor (RF) in serum as an aid in the diagnosis of Rheumatoid Arthritis (RA).

SUMMARY AND PRINCIPLES

RA is a chronic systemic disease characterized by swelling and pain in the joints and by inflammatory and degenerative processes involving cartilage, synovial membrane or muscle tissue. Early therapy helps in minimizing irreversible damage to the joints and prompt diagnosis is crucial. A characteristic of RA is the presence in the blood and in synovial fluid of a reactive group of proteins collectively known as the RF[1,2]. These are macroglobulins having a molecular weight of about one million. In the opinion of many investigators[3] the RF are antibodies directed against "altered" human gamma globulin[4-6]. The RF are found in 70-100% of definite RA cases. The occurrence of RF in osteoarthritis or rheumatic fever is less than 2% and 3% respectively. It should be noted that incidence of RF had been reported in non-rheumatic diseases such as pulmonary tuberculosis, bacterial endocarditis, syphilis, as well as others. The principle of this test is based on the immunologic reaction between RF in serum with the IgG coated onto latex particles resulting in visible agglutination.[7]

MATERIALS SUPPLIED

RF Latex Reagent: Latex particles with human IgG in a stabilized buffer with < 0.1% sodium azide.
RF Positive Control: Human serum (> 8 IU/ml RF) with < 0.1% sodium azide.
RF Negative Control: Human serum with < 0.1% sodium azide.
Glycine-Saline Buffer Concentrate (20X): To be diluted 1:20 with distilled water.
Disposable pipettes and test slides.
Additional Items Required: Serological pipettes, 12 x 75 mm test tubes and timing device.

STORAGE & STABILITY

Store reagents and controls at 2 - 8 degree Celsius. DO NOT FREEZE. Prior to use, warm reagents and controls to room temperature. Product instability is shown by improper reaction between latex reagent with the positive and negative control sera.

PRECAUTIONS

This product is for In Vitro Diagnostic Use Only. Each donor unit used in this product has been tested by an FDA approved method and found to be non-reactive for the presence of HbsAg and antibody to HIV Virus. Because no known test method can offer complete assurance that hepatitis B virus, HIV Virus, or other infectious agents are absent, all human blood based products should be handled in accordance with good laboratory practices. The preservative sodium azide may react with metal plumbing to form explosive metal oxides. In disposal, flush with a large volume of water to prevent metal azide build up.

SPECIMEN COLLECTION

The test should be performed on fresh serum specimens only. Plasma should not be used because fibrinogen may cause nonspecific agglutination of the latex particles. Fresh specimens (less than 24 hours) should be used in performing the test. If testing is delayed, specimens should be refrigerated (or frozen where applicable). Bacterial contamination may cause protein denaturation and false positive agglutination.

PROCEDURE

A. Method I (Qualitative)

1. Bring all test reagents and serum specimens to room temperature.
2. Gently shake the RF latex vial to suspend latex particles (do not use vortex mixer). Positive and negative controls should be tested with each series of test.
3. Place one drop of test serum (with disposable pipette) onto a circle on the slide. Use new pipette for each test serum. Deliver one drop of RF Latex to each circle that contains specimens or controls. Use paddle end of pipette to spread the mixture on each circle. Do not use the same paddle end to mix each mixture as this will cause cross-contamination.
4. Gently tilt and rotate slide by hand for two (2) minutes. Observe for macroscopic clumping using the indirect oblique light source.

B. Method II (Semi-Quantitative)

1. For each test serum to be titrated, set up a least 6 test tubes (12 x 75 mm) and label1:2, 1:4, 1:8, 1:16, 1:32, 1:64, etc.
2. To each tube add 0.2 ml of Diluted Glycine-Saline Buffer.
3. To Tube 1 add 0.2 ml of undiluted test serum.
4. Make two-fold serial dilutions by mixing contents of Tube 1 with pipette and transfer 0.2 ml to Tube 2. Repeat serial transfers for each tube. For the 6 tubes, the dilutions range from 1:2 to 1:64. Additional dilutions can be made. Repeat steps 3 to 7 as given in Method I (Qualitative).

RESULTS

Qualitative

Positive Result: Agglutination **Negative Result:** Smooth milky suspension

Since negative results may be caused by antigen excess, the test should be repeated using a diluted serum sample in case prozone effect is suspected.

Semi-Quantitative

Positive sera should be retested to provide verification for borderline interpretations. The hishest dilution of sample showing agglutination is the endpoint. Multiplication of the dilution factor by 8 IU/ml will yield the approximate RF level (table is only a guide to assist in the interpretation).

Dilutions/Concentration (IU/ml)

1:1 (neat specimen) = 8, 1:2 = 16, 1:4 =32, 1:8 = 64, 1:16 = 128, 1:32 = 256

QUALITY CONTROL PROCEDURE

Positive and Negative controls should be included in each test series. Positive control should produce agglutination; and Negative control should produce no agglutination.

LIMITATIONS OF THE PROCEDURE

The results of this test SHOULD NOT be used as a single diagnostic tool to make a clinical diagnosis. Instead, other clinical findings must be evaluated with other observed symptoms to aid in the final diagnosis. This test is to be performed by hand rotation. The use of a mechanical rotator could yield false positive/negative results. Strength of agglutination in screening test is not indicative of actual RF titer. Reaction time longer than 2 minutes may produce apparent false positive reactions due to a drying effect. Lipemic, haematic or contaminated sera can cause false positive reactions. Only serum should be used in this test.

EXPECTED VALUES

The clinical significance of RF lies in differentiating between RA and rheumatic fever. RF was found in about 80% of RA patient and is almost always absent in rheumatoid fever[8]. About 3.5% of known rheumatoid patients do not react in the screening test. But about 2% of sera from healthy individuals gave a positive RF reaction.

PERFORMANCE CHARACTERISTICS

Analytical sensitivity:8 (6-16) IU/ml. The performance of the RF TEST was observed to have a sensitivity of 98% and a specificity of 97%. No prozone effect was observed up to 1500IU/ml.

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