



# WAALER - ROSE TEST

Slide passive hemagglutination test  
(Without dilution of the sample)

Cat.No. 101-0219

Size 100 tests

## PRINCIPLE:

The Waaler-Rose (WR) reagent is a suspension of stabilized sheep red cells sensitized with anti-sheep rabbit IgG. The WR test reagent sensitivity has been adjusted to detect a minimum of 6 IU/mL of rheumatoid factors according with the WHO International Standard without previous sample dilution.

## SAMPLE:

Use fresh serum obtained by centrifugation of clotted blood. The sample may be stored at +2 °C to +8 °C for 48 hours before performing the test. For longer periods of time the serum must be frozen. Hemolytic, lipemic or contaminated sera must be discarded.

## REAGENTS:

- Reagent 1; White cap  
Stabilized sheep red cells in suspension  
Sodium azide 0.1%
- Positive control (Human serum); Red cap  
Rheumatoid factor (RF)\*  $\geq 6$  IU/mL  
Sodium azide 0.1%
- Negative control (Human serum); Blue cap  
Rheumatoid factors (RF)\*  $< 6$  IU/mL  
Sodium azide 0.1%

\* The human sera used in the controls have been tested and found negative for HBsAg and HIV. However a careful handling is always recommended.

## PREPARATION OF REAGENTS:

Shake the Reagent 1 (red cells) before use. After that it must be uniform and without visible clumping. The sensitivity of the test depends of the drop volume (50  $\mu$ l). Do not use droppers than those provided and hold the dropper perpendicular to the slide surface. The reagent and controls have to be stored at +2 °C to +8 °C. Do not freeze. In these conditions the components will remain stable until the expiry date printed on the label.

## PROCEDURE:

### 1. QUALITATIVE DETERMINATION

Before using the kit, allow the components to reach room temperature. Gently shake the reagent to disperse the particles. Check the reagent against the positive and negative controls in the same way as the undiluted serum.

|  |                     |
|--|---------------------|
| Serum  | 1 drop (50 $\mu$ l) |
| Reagent 1  | 1 drop              |
| Place a drop of UNDILUTED serum onto a circle of the slide. Add a drop of the Reagent 1 (red cells) next to the drop of serum.   |                     |
| Mix both drops spreading them over the full surface of the circle. Let the slide to stay on a flat surface for two minutes. After this time twist the slide 45 degrees once and let again to stay for one minute. Read the presence or absence of visible agglutination in this period of time. Unespecific agglutination could appear if the test is read later than this time. |                     |

## INTERPRETATION OF RESULTS:

A agglutination indicates a content of RF in the sample equal or greater than 6 IU/ml. The lack of agglutination indicates a RF level lower than 6 IU/ml in the sample

## 2. SEMIQUANTITATIVE TEST

This test is performed in the same way as the qualitative test but using previous dilution of the serum sample in saline (NaCl 9 g/l), PBS or Glycine buffer.

|  |             |                  |                  |             |     |
|--|-------------|------------------|------------------|-------------|-----|
| Dilutions  | 1/2         | 1/4              | 1/8              | 1/16        | ... |
| Sample serum   | 100 $\mu$ l | ...              | ...              | ...         | ... |
| Saline   | 100 $\mu$ l | 100 $\mu$ l      | 100 $\mu$ l      | 100 $\mu$ l | ... |
|  | →           | 100 $\mu$ l<br>→ | 100 $\mu$ l<br>→ | 100 $\mu$ l |     |
| Volume of sample   | 50 $\mu$ l  | 50 $\mu$ l       | 50 $\mu$ l       | 50 $\mu$ l  |     |
| Concentration will be the reciprocal of positive reading dilution: |             |                  |                  |             |     |
| 6x <sup>n</sup> ° of dilution                                      | 6 x 2       | 6 x 4            | 6 x 8            | 6 x 16      | ... |
| IU/ml  | 12          | 24               | 48               | 96          | ... |

## EXPECTED VALUES:

Adults:  $< 6$  UI/ml

## NOTE:

The Rheumatoid Factors are immunoglobulins (most of the IgM) with antibody activity. These factors are present in most of patients suffering Rheumatoid Arthritis. There are different Rheumatoid factors and it does not exit any test capable to detect all of them, because some of them acts against human IgG, other against animal IgG, and other against both IgG. We recommend the use of RF latex test, specific for detection of rheumatoid factors acting against human IgG.

## REFERENCES:

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- Adams, L.E., Hess, E.J. Amer. Technol. 48 (1978).
- Normausell, D. Immunochemistry 9, (1972).
- Dito, W. Am.Soc.Clin.Pat. 69, (1976).