LORNE LABORATORIES LTD.

GREAT BRITAIN

RAPID LATEX KIT

DIRECTIONS FOR USE

ROSE WAALER: For Detection Of Rheumatoid Factors (RF).

SUMMARY

Rheumatoid factors are a group of antibodies directed to determinants in the Fc portion of the IgG molecule. Although rheumatoid factors are found in a number of rheumatoid disorders, such as systemic lupus erythematosis (sLE) and Sjögren's syndrome, as well as in non-rheumatoid conditions, it's central role lies in aiding in the diagnosis of rheumatoid arthritis

PRINCIPLE

When used by recommended techniques, sensitised sheep erythrocytes in reagent will agglutinate (clump) in presence of rheumatoid factor (RF). No agglutination generally indicates absence of RF (see **Limitations**).

KIT DESCRIPTION

Lorne's Rose Waaler Kit is for the detection of rheumatoid factor. The reagent is a suspension of stabilised sheep erythrocytes sensitised with rabbit anti-sheep erythrocyte IgG, which agglutinate in the presence of Rheumatoid Factor (RF). All latex reagents are supplied at optimal dilution for use with all recommended techniques without the need for further dilution or addition. For lot reference number and expiry date see **Vial Labels**.

STORAGE

Do not freeze. Reagent vials should be stored at 2 - 8ºC on receipt. Prolonged storage at temperatures outside this range may result in accelerated loss of reagent reactivity.

SPECIMEN COLLECTION

Specimens should be drawn with or without anticoagulant using an aseptic phlebotomy technique. If testing is delayed specimens can be stored at 2-8ºC for 7 days or for up to 3 months at or below –20ºC. Specimens must be free from bacterial contamination, fibrin, gross lipaemia and gross haemolysis.

PRECAUTIONS

1. The kit is for *in vitro* diagnostic use only.
2. Do not use kit past expiration date (see **Vial and Box Labels**).
3. Protective clothing should be worn when handling the reagents, such as disposable gloves and a laboratory coat.
4. All the reagents must be allowed to reach 18-25ºC before use.
5. Materials used to produce the kit were tested at source and found to be negative for HBsAg, HCV and HIV 1+2 using approved microbiological tests. However, no known tests can guarantee that products derived from human or animal sources are free from infectious agents. Care must be taken in the use and disposal of each vial and its contents.

DISPOSAL OF KIT REAGENT AND DEALING WITH SPILLAGES

For information on disposal of kit reagent and decontamination of a spillage site see **Material Safety Data Sheets**, available on request.

CONTROLS AND ADVICE

1. It is recommended the RA Positive and Negative Controls be tested in parallel with each batch of tests. Tests must be considered invalid if controls do not show expected results.
2. All the reagents must be allowed to reach 18-25ºC before use.
3. Shake the reagents well before use to ensure homogeneity.
4. Do not interchange components between different kits.
5. Use of kit and interpretation of results must be carried out by properly trained and qualified personnel in accordance with the requirements of country where kit is in use. The user must determine the suitability of the kit for use in other techniques.
6. Results obtained with the Waaler Rose method do not compare with those obtained with an RA Latex test. Differences in the results between methods do not reflect differences in the ability to detect rheumatoid factors.

KIT COMPONENTS SUPPLIED

1) Rose Waaler reagent (2.5 mL): Stabilised sheep erythrocytes sensitised with rabbit anti-sheep erythrocyte IgG, pH, 8.2, and a preservative.
2) RF Positive Control (Red cap, 1 mL): Human serum with a RF concentration > 30 IU/mL and a preservative.
3) RF Negative Control (Blue cap, 1 mL): Animal serum and a preservative.
4) Pipette-Stirrers.
5) Reusable Agglutination Slide.

AADITIONAL MATERIALS AND EQUIPMENT REQUIRED

a) Glass Test Tubes (10 x 75 mm or 12 x 75 mm).
b) Pasteur and Graduated Pipettes.

RECOMMENDED QUALITATIVE TECHNIQUE

1. Allow the reagents and samples to reach room temperature. The sensitivity of the test may be reduced at low temperatures.
2. Place 50 µL of the sample and one drop of each Positive and Negative controls into separate circles on the slide test.
3. Swirl the RW reagent gently before using and add one drop (50 µL) next to the sample to be tested.
4. Mix the drops with a stirrer, spreading them over the entire surface of the circle. Use different stirrers for each sample.
5. Let the slide sit undisturbed on a flat surface for 2 minutes.
6. After the 2 minutes, very carefully tilt the slide to about a 45° angle from the horizontal and after tilting the slide let the slide sit undisturbed on a flat surface for 1 minute.
7. After the 1 minute, examine the slide macroscopically for any visible signs of agglutination.

INTERPRETATION OF QUALITATIVE RESULTS

1. **Positive:** Visible agglutination of the sensitised erythrocytes constitutes a positive result and within the accepted limitations of the test procedure, indicates a level of RF in the specimen > 8 IU/mL.
2. **Negative:** No visible agglutination of the sensitised erythrocytes constitutes a negative result and within accepted limitations of test procedure, indicates level of < 8 IU/mL RF in specimen.

RECOMMENDED SEMI-QUANTITATIVE TECHNIQUE

1. The semi-quantitative test can be performed in the same way as the qualitative test using dilutions of the serum.
2. Make doubling dilutions of serum specimen in 9 g/L saline as follows:

Dilution	Serum	Saline
1/2	100 µl undiluted serum	100 µl
1/4	100 µl 1/2 diluted serum	100 µl
1/8	100 µl 1/4 diluted serum	100 µl
1/16	100 µl 1/8 diluted serum	100 µl

3. Test the specimen dilutions in the same way as for the quantitative technique above.
4. Agglutination of the sera indicates:

Dilution	RA Levels (I.U/mL)
1/2	16 (8 x 2)
1/4	32 (8 x 4)
1/8	64 (8 x 8)
1/16	128 (8 x 16)

5. Normal levels of RF in adults is < 8 IU/mL.

RESULTS

The titre is expressed as the reciprocal of the highest dilution showing macroscopic agglutination: e.g. if this occurs in dilution 1/8, the titre is (8 * 8 I.U/mL) = 64.

STABILITY OF THE REACTIONS

Slide tests should be interpreted straight after the 1-minute period to avoid the possibility that a negative result may be incorrectly interpreted as positive due to drying of the reagent.

LIMITATIONS

1. The incidence of false positive results is about 3-5 %. Individuals suffering from infectious mononucleosis, hepatitis, syphilis as well as elderly people may give positive results.
2. Diagnosis should not be solely based on the results of the Rose Waaler method but also should be complemented with a RA test along with the clinical examination.
3. Hemoglobin (≤ 10 g/L), bilirubin (≤ 20 mg/dL) and lipemia (≤ 10 g/L), do not interfere. Other substances may interfere[3].
4. False positive or false negative results may also occur due to:
   a) Contamination of test materials
   b) Improper storage of test materials or omission of reagents
   c) Deviation from the recommended techniques

SPECIFIC PERFORMANCE CHARACTERISTICS

1. The kit has been characterised by all the procedures mentioned in the **Recommended Techniques**.
2. The Waaler Rose sensitivity is calibrated against the RF International Calibrator from the WHO (WHO 64/1 Rheumatoid Arthritis Serum).
3. Analytical sensitivity: 8 (6-16) IU/mL, under the described assay conditions.
4. Prozone effect: No prozone effect was detected up to 800 IU/mL.
5. Diagnostic sensitivity: 100 %.
6. Diagnostic specificity: 93.6 %.

DISCLAIMER

1. The user is responsible for the performance of the kit by any method other than those mentioned in the **Recommended Techniques**.
2. Any deviations should be validated prior to use using established laboratory procedures.

BIBLIOGRAPHY

1. Robert W Dorner et al. Clinica Chimica Acta 1987; 167:1-21.
2. Frederick Wolfe et al. Arthritis and Rheumatism 1991; 34: 951-960.
3. Young DS. Effects of drugs on clinical laboratory test, 4th ed. AACC Press, 1995.
4. Koritz T N et al. Journal of Immunological Methods 1980; 32:1-9.
5. Assameh S N et al. Journal of Immunological Methods 1980; 34:205-215.
6. Robert H.Shmerling et al. The American Journal of Medicine 1991; 91:528-534.

AVAILABLE KIT SIZES

Kit Size	Catalogue Number
50 Tests Per Kit	156050A

For the availability of other sizes, please contact:

Lorne Laboratories Limited
Unit 1 Cutbush Park Industrial Estate
Danehill
Lower Earley
Berkshire, RG6 4UT
England
Tel: +44 (0) 118 921 2264
Fax: +44 (0) 118 986 4518
E-mail: info@lornelabs.com

TABLE OF SYMBOLS

LOT	Batch Number	IVD	*In-vitro* Diagnostic
REF	Catalogue Reference		Store At
	Expiry Date		Manufacturer
	Read Pack Insert		