RPR TEST KIT

Catalogue Number	Product description
RPR/010	RPR Test Kit 100 T
RPR/012	RPR Test Kit 500 T

INTENDED USE
The Plasmatec RPR test kit is a non-treponemal test system for detecting syphilis using plasma or either unheated or heated serum samples.

WARNINGS AND PRECAUTIONS
For *in vitro* diagnostic use only
For professional use only

Health and Safety warnings:
All patient samples and reagents should be treated as potentially infectious and the user must wear protective gloves, eye protection and laboratory coats when performing the test.
Non disposable apparatus must be sterilised after use by an appropriate method.
Disposable apparatus must be treated as biohazardous waste and autoclaved or incinerated.
Spillages of potentially infectious material should be absorbed and disposed of as above. The site of spillage must be sterilised with disinfectant or 70% alcohol.
Do not pipette by mouth.
The test reagent is a modified form of VDRL antigen containing microparticles. Control reagents contain human serum. The human serum used has been tested and found to be negative for HIV and Hbs Ag and has also been heat treated. Nonetheless the reagent must be treated as potentially infectious and appropriate precautions should be taken when handling and on disposal. The product also contains aqueous buffer salts including sodium azide as preservative- see material safety data sheet

Analytical precautions:
Do not modify the test procedure.
Do not dilute or modify the reagents in any way.
Allow all reagents and samples to reach room temperature (18 to 30ºC) before use.
Resuspend test and control cells gently but thoroughly.
Do not interchange reagents from different kit batches.

COMPOSITION
Kit Contents
Antigen (2ml for the 100 test kit; 10ml for the 500 test kit)
This reagent is ready for use and is supplied in 2ml/10ml capped vials. Time must be allowed for the antigen to reach room temperature prior to testing and should be *WELL SHAKEN* to ensure homogeneity.
Positive/negative controls sera
Controls are supplied so the validity of the test can be checked periodically. The controls are supplied ready for use.
(1x3ml bottle and 1 dispensing needle)
These components are designed for dispensing the RPR test antigen. For use, attach the needle to the end of the bottle and draw the *WELL SHAKEN* antigen into the bottle. Expel a drop or two of antigen to eliminate the possibility of an inadequate amount of antigen being added to the sample due to the presence of air in the needle. It is extremely important to maintain bottle and needle in a **vertical position** when dispensing the antigen. At the end of each day's testing the needle should be removed, rinsed with distilled water and air dried. The dispensing needle should not be wiped. Doing so may remove the silicon coating thereby allowing some antigen to adhere to the needle, which may result in insufficient antigen being delivered.
Test Cards
These cards are for use with the **PLASMATEC** RPR antigen suspension and are specially prepared, plastic coated cards. The circles of the test cards should never be touched with the fingers, as this may invalidate test results. Each test area should only be used once and then the card should be discarded or filed for future reference.
Pipette stirrers (100 or 500)
The droppers are used to transfer serum or plasma to the test card surface and one drop is equivalent to approximately 50?l. In qualitative tests, a new dropper must be used for each test specimen. When transferring the sample from the collecting tube, the specimen must not be drawn up into the teat as this may cause false results.
Pack Insert

STORAGE AND SHELF LIFE
Store reagents, upright at 2-8ºC.
DO NOT FREEZE THE ANTIGEN REAGENT
Do not use reagents after the stated expiry date.
Discard reagents if they become contaminated or do not demonstrate the correct activity with controls.

MATERIALS AND EQUIPMENT REQUIRED BUT NOT PROVIDED.
Micropipettes for delivering 50 µlitres.
Automatic rotating table.

SPECIMEN
The kit is designed for use with either plasma or heated and unheated serum. The samples should be free from haemolysis and contamination.

PROCEDURE
Principle:
Syphilis is a veneral disease caused by the spirochaete micro-organism *T.pallidum.* As the organism cannot be cultured on artificial media the diagnosis of syphilis depends on the correlation of clinical data with the detection of specific antibody by serological tests. Serological screening tests for syphilis using cardiolipin and lecithin as antigens are simple to perform but may give rise to a small proportion of false positive results because the tests use non-treponemal antigens.
The test antigen is a modified form of VDRL Antigen containing microparticulate carbon which aids the macroscopic reading of results. A reactive result is indicated by agglutination which is readily visible without the aid of a microscope. Weak- reactive results can be easily and clearly distinguished from non-reactive patterns which display a macroscopically smooth and even appearance.
Test results are obtained in 8 minutes

Qualitative method
1. Hold the teat between the thumb and forefinger. Squeeze the teat whilst inserting the dropper onto the specimen. Then release finger pressure to withdraw the sample taking care not to transfer any cellular elements.
2. Hold the dropper over a test card circle and squeeze the teat to allow one drop (50 ?l) to fall onto the card. It is important to maintain the dropper in a vertical position whilst dispensing the sample to be tested.
3. Using the broad end of the stirrer, spread the sample of the entire area of the test circle.
4. Attach the dispensing needle to the syringe. Withdraw sufficient antigen (*WELL SHAKEN*) for the number of tests performed. Maintaining the syringe in a vertical position, allow one drop to fall on each test sample. Do not restir.

Interpretation of results
Reactives display characteristic agglutination ranging from slight (Weak-reactive) to intense (Reactive). Very weak reactive results are characterised by small agglutinates around the periphery of the test area. Negatives do not exhibit this reaction and display a macroscopically smooth and even appearance.

Quantitative method
1. Dispense one drop of 0.85% saline on circles 1 to 5, of the test card using disposable pipette. Do not spread the saline.

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2. Using an accurate volumetric pipette dispense 50?l of test sample onto circle number 1.
3. Using the dropper prepare two-fold dilutions by drawing the mixture up and down the pipette 5 or 6 times. Avoid the formation of bubbles. Transfer 50ul from circle numbers 1 to 5 which now represent the following dilutions:

Circle	1	2	3	4	5
Dilution	1:2	1:4	1:8	1:16	1:32

4. Using a mixer spread the diluted sample across the entire surface of each circle starting at the highest dilution (circle number 5). Proceed to circles number 4, 3, 2 and 1 in a similar fashion.
5. Using the syringe and needle withdraw sufficient antigen (*WELL SHAKEN*) for the number of tests being performed. Maintaining the syringe in a vertical position allow one drop to fall onto each test circle. Do not restir. Rotate the RPR test card manually or using an automatic rotator for 8 minutes at 100 revolutions/minutes.

Interpretation of results

At the end of 8 minutes of rotation, inspect the test card macroscopically, in good light.

Read the test and note the last circle in the dilution series that has a positive example:

If the highest dilution tested (1:32) is Reactive, proceed with the dilutions series as follows:-

5.1 Prepare a 1:16 dilution of the test sample by adding 0.1 ml of serum or plasma to 1.5ml of 0.85% saline. (1:16 dilution). Mix well.

5.2 Dispense one drop of 0.85% saline onto circles 6, 7, 8, 9 and 10.

5.3 Dispense one drop of the 1:16 dilution (prepared in 5.1) onto circle number 6. Then proceed as in step 2. The circles 6, 7, 8, 9 and 10 now represent the following dilutions:-

Circle	6	7	8	9	10
Dilution	1:32	1.64	1.128	1.256	1.512

Complete the test by following the steps number 3 and 4.

PERFORMANCE CHARACTERISTICS AND LIMITATIONS OF THE METHOD

In common with all reagin tests the RPR test may give a small proportion of false positive results.

Such reactions can be caused by diseases such as infectious mononucleosis, leprosy, lupus erythematosus, vaccinia and virus pneumonia.

Reactive RPR test specimens should be subject to further serological studies (i.e. TPHA, FTA, and ABS) since, as with any serological testing procedure, the diagnosis of syphilis should not be made on a single reactive result.

In common with other serological tests Plasmatec RPR cannot distinguish between syphilis and other pathogenic treponemal infections, egg. Yaws. Clinical evidence should always be considered when making a diagnosis of treponemal infections.

INTERNAL QUALITY CONTROL

Positive and negative control sera are provided and should be used to verify the test

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