SUMMARY
Syphilis is a venereal disease caused byTreponema pallidum, which invades intact mucous membranes or damaged skin areas. Sexual contact is the most common way of transmitting this disease. Microorganisms multiply and spread quickly after invasion. Illness' detection and treatment in early stages, are essential to avoid severe complications as neurosyphilis, cardiovascular syphilis and congenital syphilis. Disease's diagnosis has always been confronted with the difficulty to detect the etiological agent when skin lesions are not yet observed, as well as with the lack of culture methods for microorganisms' isolation. However, certain substances called "reagins" appear in the serum of an infected individual from the disease's onset and they react with cardiolipin/lecithin/cholesterol antigen. These reactions, together with clinical signs, are thus the quickest and more useful procedure available for syphilis diagnosis.

PRINCIPLE
In the Rapid test for Plasmatic Reagins (RPR) the “reagins” present in individuals infected with T. pallidum are detected by their reaction to a cardiolipin antigen, lecithin and cholesterol adsorbed onto charcoal particles. The reaction produces a macroscopically visible agglutination, due to addition of the charcoal particles. Non-specific reactions are avoided using highly purified antigen and adding choline chloride. Thus, inactivation of the sample is not necessary.

PROVIDED REAGENTS
A. Reagent A: antigen suspension of cardiolipin 0.003%, lecithin 0.02% and cholesterol 0.09%, adsorbed in charcoal particles 0.02% specially treated in 0.01 mol/l phosphate buffer with 0.72 mol/l choline chloride, 12.5 mmol/l EDTA and appropriate preservative. 
Positive Control: reactive serum dilution. Negative Control: non-reactive serum dilution.

NON-PROVIDED REAGENTS
- Saline solution (required for semi-quantitative test) 9 g/l NaCl.

STABILITY AND STORAGE INSTRUCTIONS
Provided Reagents are stable at 2-10°C until the expiration date stated on the box. Do not freeze.

INSTRUCTIONS FOR USE
Reagent A: ready to use. Mix well before use. To dispense the reagent use the provided metal dropper or measure 17 ul with micropipette.
Positive Control: ready to use. Negative Control: ready to use.

WARNINGS
Reagents are for "in vitro" diagnostic use. Positive and Negative Controls have been tested for the surface antigen of the hepatitis B virus and antibodies against HCV and HIV 1/2, being found non-reactive. Use the reagents according to the working procedures for clinical laboratories. The reagents and samples should be discarded according to the local regulations in force.

REQUIRED MATERIAL
1- Provided
- Reaction cards
- Metal dropper to dispense the Reagent
- Disposable plastic droppers to dispense and distribute the samples
2- Non-Provided
- Mechanical rotator adjustable at 100 rpm

SAMPLE
Serum or plasma
a) Collection: obtain in the usual way. Do not inactivate.
b) Additives: In case test is performed on plasma, use heparin, EDTA, fluoride or sodium oxalate as anticoagulant.
c) Known interfering substances: hemolysis, hyperlipemia or an anticoagulant excess, may cause erroneous results.
d) Stability and storage instructions: in case they are not processed immediately, samples can be kept in refrigerator up to 7 days (2-10°C). Plasma can be used up to 24 hours after collection.

PROCEDURE
I- QUALITATIVE TECHNIQUE
Before starting the test, bring the reagents and samples to room temperature. On each circle of the reaction card, place with one provided plastic dropper:

Sample or Controls 1 drop (50 ul)

Spread the sample evenly on the entire circle. With the provided metal dropper in vertical position, add in the
center of the circle:

Reagent A 1 drop (17 ul)

Without mixing, horizontally rotate the reaction card manually or with a mechanical rotator at 100 rpm for 8 minutes. Observe the presence or absence of agglutination after this period. Delayed reading times may yield erroneous results.

II- SEMI-QUANTITATIVE TECHNIQUE
Perform serial dilutions of 1:2; 1:4; 1:8, up to 1:64 with saline solution and proceed as described in I. The titer will be given by the inverse of the last reactive dilution observed.

INTERPRETATION OF RESULTS
Reactive: presence of visible agglutination as black clumps against a light background, indicates presence of “reagins” in sample.
Non-reactive: an homogeneous gray aspect, indicates absence of “reagins” in sample.

QUALITY CONTROL METHOD
In order to control the system’s quality, process a Positive Control and a Negative Control using both in the same way as the sample.

PROCEDURE LIMITATIONS
- See Known Interfering Substances under SAMPLE.
- Avoid finger contact with the circles of the card, may cause a wrong distribution of the sample, yielding erroneous results. If sample cannot be evenly spread on the circle, it is recommended to use another card circle.
- After the metal dropper is used to dispense the Reagent A, discard the rest and rinse with distilled water.
- The Reagent A should be dispensed into the reaction card keeping the dropper in vertical position to ensure the correct drop volume.
- In the presence of high temperatures or low humidity in the environment, it is recommended to use a humidifying cover during rotation, to prevent the samples from drying.
- Falsely positive results can be observed in individuals suffering from hepatitis, influenza, brucellosis, leprosy, malaria, asthma, tuberculosis, cancer, diabetes and autoimmune diseases. These cases generally show reactions with low titer and a medical record not coincident with syphilis symptoms.
- Falsely negative results may be observed when a prozone phenomenon appears. For this reason, it is recommended to repeat the test on serum diluted 1:5 with saline to verify results. If flocculation is observed under these conditions, sample is reactive.
- In spite of the advantages of this method, the results obtained, as those from any other serological test, must be considered only as a diagnostic aid, which must be checked against the patient’s medical record.

PERFORMANCE
Statistical studies performed indicate that the method’s sensitivity and specificity are similar to the USR (Unheated Serum Reagin) test ones.

WIENER LAB. PROVIDES
Kit for 250 tests (Cat. 1853154).

REFERENCES
Symbols

The following symbols are used in packaging for Wiener lab. diagnostic reagent kits.

- CE: This product fulfills the requirements of the European Directive 98/79 EC for "in vitro" diagnostic medical devices
- EC REP: Authorized representative in the European Community
- IVD: "In vitro" diagnostic medical device
- Σ: Contains sufficient for <n> tests
- Use by
- Temperature limitation (store at)
- Do not freeze
- Biological risks
- Volume after reconstitution
- Contents
- Batch code
- Manufactured by:
- Harmful
- Corrosive / Caustic
- Irritant
- Consult instructions for use
- Calibrator
- Control
- Positive Control
- Negative Control
- Catalog number