Atlas Medical

H. pylori Antigen ELISA Test Kit

An enzyme immunoassay (ELISA) for the qualitative and quantitative detection of Helicobacter pylori (*H. pylori*) Antigen in human stool

IVD For *in vitro* diagnostic and professional use only

2°C 8°C Store at (2° to 8°C)

Σ **96 Tests**

CE

INTENDED USE

The *H. pylori* Antigen ELISA Test Kit is an enzyme immunoassay for the qualitative and quantitative detection of *H. pylori* antigen in human stool. It is intended as an aid in the diagnosis of possible *H. pylori* infection and in the follow-up of patients undergoing antimicrobial therapy.

INTRODUCTION

Helicobater pylori are Gram-negative spiral-shaped bacteria that have adapted to living in the harsh acidic conditions of the stomach. These bacteria can alter their surrounding micro-environment by reducing its acidity so they can survive .Their spherical shape facilitates penetration of the epithelial lining, where the bacteria are protected by mucus against cells of the immune system.

Infections with *H.pylori,* though harmless during childhood*, manifest* as peptic ulcers of the stomach, duodenum and of small intestine, active and chronic gastritis, as well as non-ulcer dyspepsia, in about 60% of the global adult population. The mechanism of bacterial transmission is still unknown, but is thought to be oral and/or fecal borne.

The *H. pylori* Antigen ELISA Test Kit is an immunoassay for the qualitative and quantitative detection of *H. pylori* Antigen in human stool. The test utilizes antibodies to *H. pylori* to selectively detect *H. pylori* Antigen in human stool.

PRINCIPLE OF THE TEST

The *H. pylori* Antigen ELISA Test Kit is a solid phase enzyme immunoassay based on sandwich principle for the qualitative and quantitative detection of *H. pylori* antigen in human stool. The microwell plate is coated with anti-*H. pylori* antibodies. During testing, the antigens are extracted from the specimen with extraction solution and added onto the antibodies coated microwell plate along with the enzyme- conjugated antibodies to *H. pylori*, and then incubated. If specimens contain *H. pylori* antigens, it will bind to the antibodies coated on the microwell plate and simultaneously bind to the conjugate to form immobilized antibody-H. pylori antigen-conjugate complexes. If specimens do not contain *H.pylori* antigens, the complexes will not be formed. After initial incubation, the microwell plate is washed to remove unbound materials. Substrate A and substrate B are added and then incubated to produce a blue color indicating the amount of *H. pylori* antigens present in the specimens. Sulfuric acid solution is added to the microwell plate to stop the reaction producing a color change from blue to yellow. The color intensity, which corresponds to the amount of *H. pylori* antigens present in the specimens, is measured with a microplate reader at 450/630-700 nm or 450 nm.

PRECAUTIONS

- For professional *in vitro* diagnostic use only.
- Follow the instructions for use carefully. Reliability of assay results cannot be guaranteed if there are any deviations from the instructions in this package insert.
- Wear protective clothing and disposable gloves when dealing with samples and reagents. Wash hands after operations.
- Do not use reagents beyond the labeled expiry date.
- Do not mix or use components from kits with different batch codes.
- It is important to calibrate all the equipment e.g. micropipettes, and/or the automated instruments used with this device, and to perform routine preventative maintenance.
- Ensure that the bottom of the plate is clean and dry and that no bubbles are present on the surface of the liquid before reading the plate.
- Never eat, drink, smoke, or apply cosmetics in the assay laboratory. Never pipette solutions by mouth.
- Avoid cross contamination between reagents to ensure valid test results.
- Follow the wash procedure to ensure optimum assay performance.
- Use Plate Sealer to cover microwell plate during incubation to minimize evaporation.
- Use a new pipette tip for each specimen assayed.
- Do not touch or splash the rim of the well. Do not blow out from micropipettes.
- Do not allow sodium hypochlorite fumes from chlorine bleach or other sources to contact the microwell plate during the assay as the color reaction may be inhibited.

HEALTH AND SAFETY INFORMATION

- Collect samples in accordance with correct medical practices.
- Some reagents may cause toxicity, irritation, burns or have a carcinogenic effect as raw materials. Contact with the skin and the mucosa should be avoided but not limited to the following reagents: Stop solution, the Conjugate, and the Wash buffer, Extraction solution, Substrate.
- The Stop solution 0.5M H2SO4 is an acid. Use it with appropriate care. Wipe up spills immediately and wash with water if it comes into contact with the skin or eyes.
- ProClinTM 300 0.1% is used as a preservative; it can cause irritation of the skin. Wipe up spills immediately or wash with water if it comes into contact with the skin or eyes.
- Pipette tips, vials, strips and specimen containers should be collected and autoclaved for not less than 2 hours at 121°C before any further steps of disposal. Solutions containing sodium hypochlorite should NEVER be autoclaved.
- All specimens and materials should be considered as potentially infectious. Strict adherence to GLP (Good Laboratory Practice) regulations can ensure personal safety.
- Chemicals should be handled and disposed of only in accordance with the current GLP (Good Laboratory Practices) and the local or national regulations.
- Neutralized acids and other liquids should be decontaminated by adding sufficient volume of sodium hypochlorite to obtain a final concentration of at least 1.0%. A 30 minute exposure to 1.0% sodium hypochlorite may be necessary to ensure effective decontamination.

STORAGE AND STABILITY

- Components of the kit will remain stable through the expiration date indicated on the label and package when stored between 2-8°C, Once opened; all reagents are stable for up to 3 months after the first opening date if stored between 2-8°C. Return reagents to 2-8°C immediately after use.
- Place unused wells in the zip-lock aluminum foiled pouch and return to 2-8 °C, under which conditions the wells will remain stable for 3 months from the opening date.
- Concentrated Wash Buffer may be stored at room temperature to avoid crystallization. If crystals are present, warm up the solution at 37°C. Working Wash Buffer is stable for 2 weeks at room temperature.
- Do not expose reagents especially the Substrate to strong light or hypochlorite fumes during storage or incubation steps.
- Do not store Stop Solution in a shallow dish or return it to the original bottle after use.

SPECIMEN COLLECTION AND PREPARATION

- This *H. pylori* Antigen ELISA Test can be performed using only human stool.
- Stool samples should be collected in clean containers. Samples can be stored in the refrigerator (2-8 °C) for 1-2 days prior to testing. For longer storage, the specimen must be kept frozen at -20ºC. In this case, the sample should be totally thawed and brought to room temperature before testing.
- The patient has to be asked to collect the specimen avoiding any possible contact with urine or water.
- The patient submitted to the test should not be under antibiotic or anti-bacterial treatments as this pharmaceutical therapy is known to affect *H. pylori* up to a certain extent, depending on the antibiotic used, giving rise to false interpretation.
- If specimens are to be shipped, they should be packed in compliance with local regulations covering the transportation of etiologic agents.

MATERIALS

MATERIALS PROVIDED

1. ***H.Pylori* Antigen Microwell Plate:** Microwell plate coated with anti-*H. Pylori* antibodies. **(1 plate: 96 wells/plate).**
2. ***H.pylori* Antigen Conjugate:** One red cap vial containing antibodies to *H. pylori* bound to peroxidase; Preservative: 0.1% ProClin™ 300. **(1 x 8 mL).**
3. **Concentrated Wash Buffer (25x):** One white cap bottle containing Tris-HCl buffer containing 0.1% Tween 20; Preservative: 0.1% ProClin™ 300. **(1 x 40 mL).**
4. **Extraction Solution:** One white cap bottle containing 0.9% NaCl buffer containing EDTA; Preservative: 0.1% ProClin™ 300. **(1 x100 mL).**
5. **Substrate A:** One white cap vial containing Citrate-phosphate buffer containing hydrogen peroxide; Preservative: 0.1% ProClin™ 300. **(1 x 8 mL).**
6. **Substrate B:** One blue cap vial containing Buffer containing tetramethylbenzidine (TMB); Preservative: 0.1% ProClin™ 300. **(1 x 8 mL).**
7. **Stop Solution:** One yellow cap vial containing 0.5M Sulfuric acid. **(1 x 8 mL).**

8. ***H.pylori* Antigen Calibrator 1:** One white cap vial containing Buffer non-reactive for *H. pylori* Antigen; Preservative: 0.1% ProClin™ 300. **(1 x 1 mL).**
9. ***H.pylori* Antigen Calibrator 2:** One white cap vial containing Buffer containing 0.1 µg/mL *H. pylori* Antigen; Preservative: 0.1% ProClin™ 300. **(1 x 1 mL).**
10. ***H.pylori* Antigen Calibrator 3:** One white cap vial containing Buffer containing 0.5 µg/mL *H. pylori* Antigen; Preservative: 0.1% ProClin™ 300. **(1 x 1 mL).**
11. ***H.pylori* Antigen Calibrator 4:** One white cap vial containing Buffer containing 1.0 µg/mL *H. pylori* Antigen; Preservative: 0.1% ProClin™ 300. **(1 x 1 mL).**
12. **Plate Sealers (2 pieces).**
13. **Package Insert (1 copy).**

MATERIALS REQUIRED BUT NOT PROVIDED

- Freshly distilled or deionized water.
- Sodium hypochlorite solution for decontamination.
- Absorbent paper or paper towel.
- Water bath or incubator capable of maintaining 15°C to 30°C.
- Calibrated automatic or manual microwell plate washer capable of aspirating and dispensing 350 µL/well.
- Disposable gloves.
- Automated processor (optional).
- Calibrated micropipettes with disposable tips capable of dispensing 50 and 100 µL.
- Graduated cylinders for wash buffer dilution.
- Vortex mixer for specimen mixing (optional).
- Disposable reagent reservoirs.
- Calibrated microplate reader capable of reading at 450 nm with a 630-700 nm reference filter, or reading at 450 nm without a reference filter.
- Timer.

DIRECTIONS FOR USE

1. Remove unused strips from the microwell plate, and store in the original resealable pouch at 2-8°C.
2. Allow reagents and specimens to reach room temperature (15-30°C) prior to testing.

WASH PROCEDURE

a. The wash procedure is critical. Insufficient washing will Result in a poor precision and falsely elevated absorbance readings.

b. Prepare working wash buffer by adding content of wash buffer bottle provided with the kit to distilled or deionized water to reach a final volume of 1 liter. The working wash buffer is stable for 2 weeks at 15-30°C.

3. Dispense 1 mL of Extraction Solution into Specimen Extraction Tube.

For Solid Stool Specimens:

i. Take out the cap of the Specimen Extraction Tube
ii. Randomly stab the specimen collection stick into the stool specimen in at least 3 different sites to collect approximately 30 mg of specimen (equivalent to 1/4 of a pea). Do not scoop the stool specimen.
iii. Transfer into Specimen Extraction Tube.

For Liquid Stool Specimens:

i. Hold the Liquid Specimen Dropper vertically.
ii. Aspirate stool specimens and then dispense 2 drops (approximately 50 µL) into the Specimen Extraction Tube containing the Extraction Solution.
iii. Screw on and tighten the cap onto the Specimen Extraction Tube.
iv. Shake the Specimen Extraction Tube vigorously to mix the specimen and the Extraction Solution.

4. Leave A1 as Blank well.
5. Dispense 50 µL of Calibrator 1 in wells B1 and C1. (Light Yellow Reagent)
6. Dispense 50 µL of Calibrator 2 in wells D1 and E1. (Green Blue Reagent)
7. Dispense 50 µL of Calibrator 3 in wells F1 and G1. (Light Blue Reagent)
8. Dispense 50 µL of Calibrator 4 in wells H1 and A2. (Dark Blue Reagent)
9. Hold the Specimen Extraction Tube upright and break off the tip of the tube. Invert the Specimen Extraction Tube and dispense 2 drops of the specimen Extraction Solution (approx. 50 µL) to assigned wells starting at B2. (Yellow Reagent)
10. Dispense 50 µL of Conjugate to each well except for the Blank well. (Red Reagent)
11. Mix gently by swirling the microwell plate on a flat bench for 30 seconds.
12. Cover the microwell plate with the Plate Sealer and incubate at room temperature (15-30°C) in a room, a water bath, or an incubator for 60 minutes ± 5 minutes.
13. Remove the Plate Sealer.
14. Wash each well 5 times with 350 µL of Working Wash Buffer per well, and then remove the liquid.
15. Turn the microwell plate upside down on absorbent tissue for a few seconds. Ensure that all wells have been completely washed and dried.
    Note: Improper washing may cause false positive results.
16. Dispense 50 µL of Substrate A to each well. (Clear Reagent)
17. Dispense 50 µL of Substrate B to each well. (Clear Reagent) Then a blue color should develop in wells containing Positive specimens.
18. Mix gently then cover microwell plate with Plate Sealer and incubate at room temperature (15-30°C) in a room, a water bath, or an incubator for 10 minutes ± 1 minute.
19. Remove the Plate Sealer.
20. Dispense 50 µL of Stop Solution to each well. (Clear Reagent) Then a yellow color should develop in wells containing Positive specimens.
21. Read at 450/630-700 nm within 30 minutes.
22. Note: Microwell plate can also be read at 450 nm, but it is strongly recommended to read it at 450/630-700 nm for better results.

VALIDATION REQUIREMENT AND QUALITY CONTROL

1. Calculate the Mean Absorbance of Calibrators 1-4 by referring to the table below.

Example of Calibrator 2 Calculation

Item	Absorbent
Calibrator 2: Well D1	0.469
Calibrator 2: Well E1	0.507
Total Absorbance of Calibrator 2	0.976
Mean Absorbance of Calibrator 2	0.488

2. Check the validation requirements below to determine if the test results are valid.

Item	Validation Requirements
Blank Well	Blank Absorbance should be < 0.050 if read at 450/630-700 nm Note: It should be < 0.100 if read at 450 nm
Calibrator 1	Mean Absorbance after subtraction of Blank Absorbance should be < 0.100
Calibrator 2	Mean Absorbance after subtraction of Blank Absorbance should be > 0.150
Calibrator 3	Mean Absorbance after subtraction of Blank Absorbance should be > 0.500
Calibrator 4	Mean Absorbance after subtraction of Blank Absorbance should be > 1.000

NOTE: The test results are considered invalid if the above validation requirements are not met. Repeat the test or contact your local distributor.

INTERPRETATION OF RESULTS

Qualitative

Calculate the Index Value to obtain qualitative specimen results.

1. If the test is valid, obtain Cut-Off Value by subtracting the Blank Absorbance from the Mean Absorbance of 1/2× (Calibrator 2+Calibrator 1). See an example of Cut-Off Value calculation below.

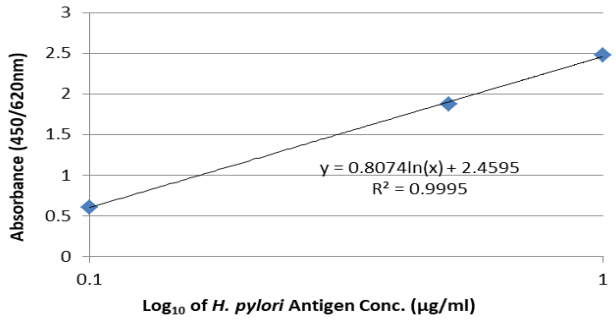
Item	Absorbent
Blank Absorbance: Well A1	0.011
Cut-Off Value: 1/2× (Mean Absorbance of Calibrator 2+ Mean Absorbance of Calibrator 1) – Blank Absorbance	1/2× (0.488+0.012)-0.011=0.239

2. Calculate the Index Value by dividing the Specimen Absorbance by the Cut-Off Value, and then read the results by referring to the Interpretation of Results table below.

Item	Absorbent
Specimen: Well F2	0.968
Blank Absorbance: Well A1	0.011
Cut-Off Value	0.239
Index Value: Specimen/Cut-Off Value	(0.968-0.011)/0.239=4.0

Quantitative

Draw the calibration curve and obtain quantitative specimen results.



1. Subtract the Blank Absorbance from the Mean Absorbance of each Calibrator, and then plot them on the Y-axis against their Log_{10} of the corresponding concentration in µg/mL on the X-axis on a linear graph paper and draw the calibration curve. Draw the best fitted line through data points to obtain a standard curve. Refer to an example of the calibration curve at right.

   NOTE: Do not use the calibration curve at right to make any calculation. A calibration curve must be performed for each run.
2. Obtain quantitative specimen results from their absorbance by using the calibration curve.

   NOTE: Specimens that have absorbance above Calibrator 4 should be pre-diluted using Extraction Solution and retested. The concentration must be multiplied by the dilution factor. Automated reading and calculation may also be performed using linear regression function on suitable computer programs.

Interpretation of Results - Qualitative and Quantitative

Results	Qualitative	Quantitative
	Index Value	Concentration
Negative	< 0.9	< 0.045 µg/mL
Positive	> 1.1	> 0.055 µg /mL
Equivocal*	≥ 0.9 and ≤ 1.1	0.045 – 0.055 µg/mL

***NOTE:** For Equivocal results, the specimen should be retested. Specimens that are repeatedly Equivocal after retest should be confirmed using an alternate method. If the results remain Equivocal, collect a new specimen in two weeks. If the new specimen is Positive, the specimen is presumed to be Positive.

LIMITATIONS

1. The *H. pylori* Antigen ELISA Test Kit is used for the detection of *H. pylori* antigen in human stool. Diagnosis of an infectious disease should not be established based on a single test result. Further testing, including confirmatory testing, should be performed before a specimen is considered positive. A negative test result does not exclude the possibility of exposure. Specimens containing precipitate may give inconsistent test results.
2. As with all diagnostic tests, all results must be interpreted together with other clinical information available to the physician.
3. As with other sensitive immunoassays, a false positive result may arise due to inadequate washing from the initial test. The results may be affected due to procedural or instrument error.

PERFORMANCE CHARACTERISTICS

Sensitivity and Specificity

The *H. pylori* Antigen ELISA Test Kit has been compared to a leading commercial *H. Pylori* Antigen ELISA test using clinical specimens. The results show that the clinical sensitivity of the *H. pylori* Antigen ELISA Test Kit is 98.6%, and the clinical specificity is 95.4%.

***H. pylori* Antigen ELISA vs. Other ELISA**

Method		Other ELISA		Total Results
H. pylori Antigen ELISA	Results	Positive	Negative	
	Positive	70	6	76
	Negative	1	125	126
Total Results		71	131	202

Clinical Sensitivity: 98.6% (92.4-100.0%) *
Clinical Specificity: 95.4% (90.3-98.3%)*
Overall Agreement: 96.5% (93.0-98.6%)*
***95% Confidence Interval**

Reproducibility

Intra-Assay: Within-run precision has been determined by using 10 replicates of two specimens: a low positive and a high positive.

Inter-Assay: Between-run precision has been determined by using 10 replicates on the same two specimens: a low positive and a high positive. Three different lots of the *H. pylori* Antigen ELISA Test Kit have been tested using these specimens.

Specimen	Intra-Assay			Inter-Assay		
	Mean Absorbance/ Cut-Off	Standard Deviation	Coefficient of Variation (%)	Mean Absorbance/ Cut-Off	Standard Deviation	Coefficient of Variation (%)
1	1.741	0.156	8.96	1.723	0.133	7.72
2	4.726	0.252	5.33	4.861	0.252	5.18

REFERENCES

1. Marshall, BJ, McGechie, DB, Rogers, PAR and Glancy, RG. Pyloric Campylobacter infection and gastroduodenal disease. Med. J. Australia. (1985), 149: 439-44.
2. Soll, AH. Pathogenesis of peptic ulcer and implications for therapy. New England J. Med. (1990), 322: 909-16.
3. Ansorg, R, Von Recklinghausen, G, Pomarius, R and Schmid, EN. Evaluation of techniques for isolation, subcultivation and preservation of *Helicobacter pylori*. J. Clin. Micro. (1991), 29:51-53.
4. Pronovost, AP, Rose, SL, Pawlak, J, Robin, H and Schneider, R. Evaluation of a new immunodiagnostic assay for *Helicobacter pylori* antibody detection: Correlation with histopathological and microbiological results. J. Clin. Micro. (1994), 32: 46-50.
5. Megraud, F, Bassens-Rabbe, MP, Denis, F, Belbouri, A and Hoa, DQ. Seroepidemiology of *Campylobacter pylori* infection in various populations. J. Clin. Micro. (1989), 27: 1870-3.

ATLAS MEDICAL
Ludwig-Erhard Ring 3
15827 Blankenfelde-Mahlow
Germany
Tel: +49 - 33708 – 3550 30
Email: Info@atlas-medical.com
Website: www.atlas-medical.com

PPI1532A01
Rev B (21.10.2021)

REF	Catalogue Number		Temperature limit
IVD	In Vitro diagnostic medical device		Caution
	Contains sufficient for <n> tests and Relative size		Consult instructions for use (IFU)
LOT	Batch code		Manufacturer
	Fragile, handle with care		Use-by date
	Manufacturer fax number		Do not use if package is damaged
	Manufacturer telephone number		Date of Manufacture
	Keep away from sunlight		Keep dry

Atlas Medical CE

ATLAS SLE SLIDE TEST

IVD For *in vitro* diagnostic and professional use only

Store at 2°-8°C

INTENDED USE

Atlas SLE Slide Test is a slide agglutination assay for the qualitative and semi quantitative detection of anti-deoxyribonucleoprotein (anti-DNP) in human serum. No initial dilution of patient samples is required for this test. These materials are intended to be acquired, possessed and used only by health professionals.

INTRODUCTION

The detection of antinuclear antibodies, by such laboratory methods as immunofluorescence, LE cell test, and agglutination of coated particles, can aid in the diagnosis of such autoimmune diseases as systemic lupus erythematosus (SLE). The antibodies most associated with SLE are those directed against DNP. These antibodies are believed to cause the formation of the LE cell *in vitro*, occurring in 75-80% of patients diagnosed as having SLE. Given that 20-25% of SLE patients do not exhibit the formation of LE cells, other methods can be used to detect antinuclear antibodies.

PRINCIPLE

Atlas SLE Slide Test provides a means of detecting anti-DNP in human serum. SLE Slide reagent is a stabilized buffered suspension of polystyrene latex particles that have been coated with DNP. When the latex reagent is mixed with the serum containing antibodies to DNP, agglutination occurs. Using dilutions of a reactive patient sample, the anti-DNP titer can be determined.

MATERIALS

MATERIALS PROVIDED

- SLE Latex Reagent: Suspended inert latex particles coated with DNP, with 0.1% sodium azide as preservative.
- SLE Positive Human serum or defibrinated plasma (liquid), with 0.1% sodium azide as preservative.
- SLE Negative Control: Non-reactive buffer containing BSA and 0.1% sodium azide.
- Stirring sticks.
- Glass slide.
- Package insert.

MATERIALS NEEDED BUT NOT PROVIDED

- Timing device.
- 13 x 75 mm test tubes
- Volumetric pipet to deliver 0.25 ml
- Saline (0.9% NaCl solution)
- Mechanical rotator (optional)

PACKAGING CONTENTS

REF **8.00.11.0.0025 (1x1 mL Latex, 1x0.5 mL Positive Control, 1x0.5 mL Negative Control)**

REF **8.00.11.0.0050 (1x2 mL Latex, 1x0.5 mL Positive Control, 1x0.5 mL Negative Control)**

REF **8.00.11.0.0100 (1x4 mL Latex, 1x1 mL Positive Control, 1x1 mL Negative Control)**

PRECAUTIONS

- For *in vitro* diagnostic use.
- Latex reagent and controls contain sodium azide. Azides in contact with lead and copper plumbing may react to form highly explosive metal azides. When disposing of reagents containing azide, flush down the drain with large quantities of water to prevent azide build-up.
- The controls contain human serum or plasma which has been tested at the donor level for HBsAg and for HIV-1, HIV-2 and HCV antibodies and found to be nonreactive. As no known test offers complete assurance that infectious agents are absent, the controls should be considered potentially infectious and universal precautions should be used.
- Do not pipet by mouth.
- Do not smoke, eat, drink or apply cosmetics in areas where plasma/serum samples are handled.
- Any cuts, abrasions or other skin lesions should be suitably protected.
- In order to obtain reliable and consistent results, the instructions in the package insert must be strictly
- followed. Do not modify the handling and storage conditions for reagents or samples.
- Do not use past the expiration date indicated on the kit.
- Do not interchange components of one kit with those of another kit.
- Turbidity or precipitation in controls is indicative of deterioration and the component should not be used.
- Bacterial contamination of reagents or specimens may cause false positive results.

STORAGE & STABILITY

- Store all reagents at 2-8°C in an upright position when not in use.
- Do not freeze reagents.

SPECIMEN COLLECTION and STORAGE

- Use only serum that is free from contamination. Test samples should not be heat-inactivated.
- It is preferable to test samples on the day of their collection. If samples cannot be tested immediately, maintain them in their original tubes at 2-8°C and test within 48 hours.
- Serum samples stored longer than 48 hours should be stored at -20°C or below until testing. Avoid repeated freezing and thawing of specimens.
- If necessary before testing, centrifuge the specimens at a force sufficient to sediment cellular components.
- Samples to be sent out for testing should be placed on ice packs and packaged like any other biohazardous material that could potentially transmit infection.

REAGENT PREPARATION

- Allow all reagents and samples to warm to room temperature (20-30°C) before use. Do not heat reagents in a water bath.
- All reagents are ready for use as supplied. Gently mix the reagents before use; avoid foaming.
- Gently mix the latex reagent before each use to ensure homogeneity.

PROCEDURES

A. Method I (Qualitative)

1. Dispense (35 µL) of each serum sample onto a separate circle on the test slide. Add one drop of Positive and negative controls from the dropper vials supplied onto a separate circle on the test slide.
2. Dispense one drop of latex reagent (35 µL) to each serum specimen and to each control.
3. Using the flat end of the stirring sticks, mix each specimen and control serum with the latex reagent, in a circular manner, over the entire area in the circles of the card.

4. Gently tilt and rotate the card for one (1) minute and observe for agglutination. All test results should be compared to both positive and negative controls.

INTERPRETATION OF RESULTS (QUALITATIVE)

Agglutination indicates a reactive SLE sample. Sera that elicit a reactive result should be retested and tittered using the "Semi quantitative Assay Protocol".

B. Method II (Semi-Quantitative)

1. Prepare serial dilutions of patient serum, in saline, in test tubes as follows:

Tube	Dilution	Composition
1	1:2	0.25 ml of serum + 0.25 ml saline.
2	1:4	0.25 ml from tube 1 + 0.25 ml saline.
3	1:8	0.25 ml from tube 2 + 0.25 ml saline.
4	1:16	0.25 ml from tube 3 + 0.25 ml saline.
5	1:32	0.25 ml from tube 4 + 0.25 ml saline.
6	1:64	0.25 ml from tube 5 + 0.25 ml saline.

Note: Testing on additional dilutions should be performed as needed.

2. Using each dilution as a separate test specimen, apply the samples to the slide as described in Step 1 of the "Qualitative method" and proceed with Steps 2 through 4 of the "Qualitative method". Include undiluted sample if not tested previously on that day with the same lot of latex reagent.

INTERPRETATION OF RESULTS (SEMI-QUANTITATIVE)

The highest dilution in which visible agglutination occurs is considered the endpoint titer.

QUALITY CONTROL

Quality Control requirements must be performed in accordance with applicable local, state and/or federal regulations or accreditation requirements and your laboratory's standard Quality Control Procedures. Controls with graded reactivity should be included. If control samples do not yield the expected response, the assay should be considered invalid and the assay repeated. If the repeat assay does not elicit the expected results for the control samples, discontinue use of the kit and contact your local distributer.

EXPECTED VALUES

Serum samples from 155 individuals were tested using the **SLE Slide Test**. Of the 155 individuals, 29 had active SLE, 23 had clinically inactive SLE, 8 had connective tissue diseases and the remaining 95 were either clinically normal or had some nonrelated disease (including anemia, infectious mononucleosis and rheumatic heart disease) and were used as controls. Results from testing with the **SLE Slide Test** were compared with the results from testing of the samples using a standard LE cell preparation assay and a fluorescent ANA assay.

Of the 29 active SLE patients, 82% were positive using the SLE Slide Test, 86% were positive by the LE cell prep, and 82% positive by the ANA test. For the 23 clinically inactive SLE patients, 19% were positive by both the SLE Slide Test SLE and the LE cell prep; and 71% were positive by the ANA test. None of the 8 patients having connective tissue disease tested positive with the SLE Latex Test, whereas 17% and 50% tested positive by the LE cell prep and the ANA procedures, respectively. Of the controls, 1% tested positive by both the SLE Latex Test and the LE cell prep, while 6% tested positive by the ANA assay.

LIMITATION

1. Serum from patients with scleroderma, rheumatoid arthritis, dermatomyositis, and a variety of connective tissue diseases may elicit agglutination in the SLE slide test.
2. Because extremely high levels of antibodies might affect the degree of agglutination, positive samples should be reassayed using the semi quantitative procedure.
3. Contaminated, lipemic, or grossly hemolyzed sera should not be used because of the possibility of nonspecific results.
4. Plasma samples should not be used because of the possibility of nonspecific results.
5. Samples yielding indeterminate results may be resolved by repeating the test utilizing a two (2) minute slide rotation period. Reaction times longer than two minutes might cause false positive results due to a drying effect.
6. Drugs such as hydralazine, isoniazid, procainamide and a number of anticonvulsant drugs can induce an SLE syndrome.
7. In accord with all diagnostic methods, a final diagnosis should not be made on the result of a single test, but should be based on a correlation of test results with other clinical findings.

REFERENCES

1. Christian CL, Mendez-Bryan R, Larson DL. 1958. *Proc Exp Biol Med*, **98**:820-823.
2. Friou GJ, Finch SC, Detre KD. 1958. *J Immunol*, **80**:324-329.
3. Hargraves MM, Richmond H, Morton R. 1948. *Proc Mayo Clin*, **23**:25-28.
4. Holman HR, Kunkel HG. 1957. *Science*, **126**:163.
5. Miescher PA, Strassel R. 1957. *Vox Sang*, **2**:283-287.
6. Miescher PA, Rothfield N, Miescher A. 1966. Lupus Erythematosus, EL Dubois, Ed., Blakiston Co., New York.
7. Rothfield NF, Phythyon JJ, McEwan C., Miescher P. 1961. *Arth Rheum*, **4**:223-229.

ATLAS Medical GmbH
Ludwig-Erhard Ring 3
15827 Blankenfelde-Mahlow
Germany
Tel: +49 - 33708 – 3550 30
Email: Info@atlas-medical.com
Website: www.atlas-medical.com

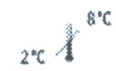
PPI2339A01
Rev B (22.06.2023)

REF	Catalogue Number		Temperature limit
IVD	*In Vitro* diagnostic medical device		Caution
Σ	Contains sufficient for <n> tests and Relative size		Consult instructions for use (IFU)
LOT	Batch code		Manufacturer
	Fragile, handle with care		Use-by date
	Manufacturer fax number		Do not use if package is damaged
	Manufacturer telephone number		Date of Manufacture
	Keep away from sunlight		Keep dry
CONTROL +	Positive control	CONTROL -	Negative control

Atlas Medical CE

RPR SYPHILIS CARD TEST

IVD *For In-Vitro diagnostic and professional use only*



Store at 2 to 8 °C

INTRODUCTION

Syphilis is a disease caused by infection with the spirochete Treponema pallidum. The infection is systemic and the disease is characterized by periods of latency. These features, together with the fact that T pallidum cannot be isolated in culture, mean that serologic techniques play a major role in the diagnosis and follow-up of treatment for syphilis.

Syphilis is categorized by an early primary infection in which patients may have non-specific symptoms, and potentially, genital lesions. Patients tested by serology during the primary phase may be negative for antibodies, especially if testing is performed during the first 1 to 2 weeks after symptom onset. As the disease progresses into the secondary phase, antibodies to T pallidum reach peak titers, and may persist indefinitely regardless of the disease state or prior therapy. Therefore, detection of antibodies to nontreponemal antigens, such as cardiolipin (a lipoidal antigen released by host cells damaged by T pallidum) may help to differentiate between active and past syphilis infection. Nontreponemal antibodies are detected by the rapid plasma reagin (RPR) assay, which is typically positive during current infection and negative following treatment or during late/latent forms of syphilis.

PRINCIPLE

RPR utilises carbon particles coated with cardiolipin antigen to detect reagin antibodies present in serum or plasma of syphilitic persons.

Specimens that contain reagin cause aggregation of the carbon particles which appear as dark clumps against a white background. The aggregation can be read macroscopically. Non-reactive samples typically appear as a smooth non-aggregated pattern which may form buttons in the centre of the test area.

MATERIALS

MATERIALS PROVIDED

- **RPR carbon antigen reagent**:Contains less than 0.1% sodium azide.
- **Positive Control** : Contains less than 0.1% sodium azide.
- **Negative control**: Contains less than 0.1% sodium azide
- **RPR test cards (Optional).**
- **Plastic sticks.**
- **Package insert.**

NOTE: This package insert is also used for individually

MATERIALS NEEDED BUT NOT PROVIDED

- Rotator (100rpm).
- Timer.
- Pipettes.

PACKED REAGENT

REF **8.00.18.0.0100 (2ml latex, 2x0.5 ml control)**

SAMPLES

Fresh serum or plasma. The samples with presence of fibrin should be centrifuged before testing. Do not use highly hemolized or lipemic samples.

PRECAUTIONS

- For professional in vitro diagnostic use only. Do not use after expiration date.
- Do not eat, drink or smoke in the area where the specimens or kits are handled.
- Always use a fresh pipette tip for every test.
- Handle all negative and positive in the manner as patient specimens .
- Wear protective clothing such as laboratory coats, disposable gloves and eye protection when specimens are assayed.
- The used test should be discarded according to local regulations.
- Components of different human origin have been tested and found to be negative for the presence of antibodies anti- HIV 1+2 and anti-HCV, as well as for HBsAg. However, the controls should be handled cautiously as potentially infectious.

STORAGE AND STABILITY

All components of the kit are stable until the expiration date on the label when stored tightly closed at 2-8°C.

PROCEDURES

QUALITATIVE PROCEDURE

- **Mix well the RPR reagent before use.**

1. Bring the reagents and samples to room temperature.
2. Dispense **50 µL of each sample** into a separate circle on the card. Use a separate tip for each sample.
3. Dispense **1 drop of each of positive and negative controls** into two additional circles.
4. Gently shake the dispensing vial and slightly press to remove air bubbles from the needle and the drop obtained is correct.
5. Dispense **1 drop (17.5 µl) of RPR antigen** to each circle next to the sample to be tested.
6. Place the card on a mechanical rotator and rotate at 100 r.p.m. for 8 minutes.
7. Observe macroscopically for agglutination within a minute after removing the card from the rotator.

SEMI-QUANTITATIVE PROCEDURE

- **Mix well the RPR reagent before use**.

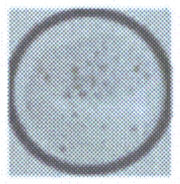
1. Make doubling dilutions from Undiluted to 1:16 normal saline.
2. Place 50 µl of each dilution in to a separate circle on the test card.
3. Spread each dilution evenly over the test circle.
4. Continue as from Qualitative procedure .
   The titer of the sample is expressed as the final dilution which shows aggregation of the carbon particles.

PERFORMANCE CHARACTERISTICS

1. **Sensitivity:** 100%.
2. **Specificity**: 100%.

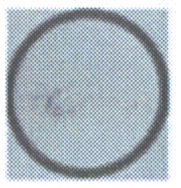
INTERPRETATION OF TEST RESULTS

1. **Strong Reactive:** Large clumps of carbon particles with a clear background.

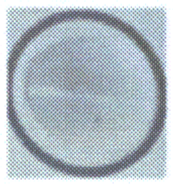


2. **Reactive:** Large clumps of carbon particles somewhat more disperse than Strong Reactive pattern.

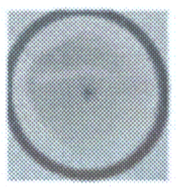
3. **Weak Reactive:** Small clumps of carbon particles with light grey background.



4. **Trace Reactive:** Slight clumping of carbon particles typically seen as a button of aggregates in the centre of the test circle or dispersed around the edge of the test circle.



5. **Non-Reactive:** Typically a smooth grey pattern or a button of non-aggregated carbon particles in the centre of the test circle.



REFERENCES

- Falcone V.H., Stout G.W. and Moore M.B. Jr., PHR 79: 491-495, 1964.

ATLAS Medical GmbH
Ludwig-Erhard Ring 3
15827 Blankenfelde-Mahlow
Germany
Tel: +49 - 33708 – 3550 30
Email: Info@atlas-medical.com
Website: www.atlas-medical.com

PPI2280A01
Rev A (10.10.2022)

REF	Catalogue Number		Temperature limit
IVD	*In Vitro* diagnostic medical device		Caution
Σ	Contains sufficient for <n> tests and Relative size		Consult instructions for use (IFU)
LOT	Batch code		Manufacturer
	Fragile, handle with care		Use-by date
	Manufacturer fax number		Do not use if package is damaged
	Manufacturer telephone number		Date of Manufacture
	Keep away from sunlight		Keep dry

Atlas Medical

ASO LATEX KIT

IVD For in -*vitro* diagnostic and professional use only

2°C 8°C Store at 2-8°C. CE

INTENDED USE

ATLAS ASO latex Test is used for the qualitative and semi-quantitative measurement of antibodies to Antistreptolysin-O in human serum.

INTRODUCTION

The group A ß-hemolytic streptococci produce various toxins that can act as antigens. One of these exotoxins streptolysin-O, was discovered by Todd in 1932.

A person infected with group A hemolytic streptococci produces specific antibodies against these exotoxins, one of which is antistreptolysin-O. The quantity of this antibody in a patient's serum will establish the degree of infection due to the hemolytic streptococcal.

The usual procedure for the determination of the antistreptolysin titer is based on the inhibitory effect that the patient's serum produces on the hemolytic power of a pre-titrated and reduced streptolysin-O. However, the antigen-antibody reaction occurs independently of the hemolytic activity of streptolysin-O. This property enables the establishment of a qualitative and quantitative test for the determination of the antistreptolysin-O by agglutination of latex particles on slide.

PRINCIPLE

ASO test method is based on an immunologic reaction between streptococcal exotoxins bound to biologically inert latex particles and streptococcal antibodies in the test sample. Visible agglutination occurs when increased antibody level is present in the test specimen.

MATERIALS

MATERIALS PROVIDED

- ASO Latex Reagent: Latex particles coated with streptolysin O, pH, 8,2. Preservative.
- ASO Positive Control (Red cap): Human serum with an ASO concentration > 200 IU/mL.Preservative.
- ASO Negative Control (Blue cap) Animal serum. Preservative
- Glass Slide.
- Stirring Sticks.

Note: This package insert is also used for individually packed reagent.

MATERIALS REQUIRED BUT NOT PROVIDED

- Mechanical rotator with adjustable speed at 80-100 r.p.m.
- Vortex mixer.
- Pippetes 50 µL.
- Glycine Buffer-20x (1000 mmol/l): add one part to nineteen parts of distilled water before use.

Packaging contents

REF **8.00.02.0.0100 (1x4ml Latex Reagent, 1x1ml positive control, 1x1ml negative control)**

PRECAUTIONS

- All reagents contain 0.1 %(w/v) sodium azide as a preservative.
- Protective clothing should be worn when handling the reagents.
- Wash hands and the test table top with water and soap once the testing is done.
- Reagents containing sodium azide may be combined with copper and lead plumbing to form highly explosive metal azides. Dispose of reagents by flushing with large amounts of water to prevent azide buildup.
- For In Vitro diagnostic use.
- Components prepared using human serum found negative for hepatitis B surface antigen (HBsAg), HCV and antibody to HIV (1/2) by FDA required test. However, handle controls as if potentially infectious.
- Accuracy of the test depends on the drop size of the latex reagent (40µl). Use only the dropper supplied with latex and hold it perpendicularly when dispensing.
- Use a clean pipette tip and stirring stick for each specimen, and glass slides should be thoroughly rinsed with water and wiped with lint-free tissue after each use.
- Check reactivity of the reagent using the controls provided.
- Do not use these reagents if the label is not available or damaged.
- Do not use the kit if damaged or the glass vials are broken or leaking and discard the contents immediately.
- Test materials and samples should be discarded properly in a biohazard container.

REAGENT PREPARATION:

The ASO Latex reagent is ready to use. No preparation is required. Mix gently before use to ensure a uniform suspension of particles.

STORAGE AND STABILITY

- Reagents are stable until specified expiry date on bottle label when stored refrigerated (2-8°C).
- **DO NOT FREEZE.**
- The ASO Latex Reagent, once shaken must be uniform without visible clumping. When stored refrigerated, a slight sedimentation may occur and should be considered normal.
- Do not use the latex reagent or controls if they become contaminated.
- Always keep vials in vertical position. If the position is changed, gently mix to dissolve aggregates that may be present.
- Reagents deterioration: Presence of particles and turbidity.

SAMPLES

- Use fresh serum collected by centrifuging clotted blood.
- If the test cannot be carried out on the same day, store the specimen for 7 days at 2-8°C and for 3 months at -20°C.
- Samples with presence of fibrin should be centrifuged before testing. Do not use highly hemolyzed or lipemic samples.
- **DO NOT USE PLASMA.**

PROCEDURE

Qualitative method

1. Allow the reagents and samples to reach room temperature. The sensitivity of the test may be reduced at low temperatures.
2. Place (40 µL) of the sample and one drop of each Positive and Negative controls into separate circles on the slide test.
3. Mix the ASO-latex reagent vigorously or on a vortex mixer before using and add one drop (40 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. Place the slide on a mechanical rotator at 80-100 r.p.m. for 2 minutes. False positive results could appear if the test is read later than two minutes.

Semi-quantitative method

1. Make serial two-fold dilutions of the sample in 9 g/L saline solution.
2. Proceed for each dilution as in the qualitative method.

QUALITY CONTROL

- Positive and Negative Controls should be included in each test batch.
- Acceptable performance is indicated when a uniform milky suspension with no agglutination is observed with the ASO Negative Control and agglutination with large aggregates is observed with the ASO Positive Control.

CALCULATIONS

The approximate ASO concentration in the patient sample is calculated as follows:

200 x ASO Titer = IU/mL

READING AND INTERPRETATION

Examine macroscopically the presence or absence of visible agglutination immediately after removing the slide from the rotator. The presence of agglutination indicates an ASO concentration equal or greater than 200 IU/mL

The titer, in the semi-quantitative method, is defined as the highest dilution showing a positive result.

REFERENCE VALUES

Up to 200 IU/mL(adults) and 100 IU/mL (children < 5 years old). Each laboratory should establish its own reference range.

PERFORMANCE CHARACTERISTICS

Analytical sensitivity:

200 (±50) IU/ml.

PROZONE EFFECT

No prozone effect was detected up to 1500 IU/ml.

SENSITIVITY

98%.

SPECIFICITY

97%.

INTERFERENCES

NON-INTERFERING SUBSTANCES:

- Hemoglobin (10 g/L)
- Bilirubin(20 mg/dL)
- Lipids (10 g/L)
- Rheumatoid factors (300 IU/mL)
- Other substances may interfere.

LIMITATIONS

- Reaction time is critical. If reaction time exceeds 2 minutes, drying of the reaction mixture may cause false positive result.
- Freezing the ASO Latex Reagent will result in spontaneous agglutination.
- Intensity of agglutination is not necessarily indicative of relative ASO concentration; therefore, screening reactions should not be graded.
- False positive results may be obtained in conditions such as, rheumatoid arthritis, scarlet fever, tonsilitis, several streptococcal infections and healthy carriers.
- Early infections and children from 6 months to 2 years may cause false negative results. A single ASO determination does not produce much information about the actual state of the disease.
- Titrations at biweekly intervals during 4 or 6 weeks are advisable to follow the disease evolution.
- Clinical diagnosis should not be made on findings of a single test result, but should integrate both clinical and laboratory data.

REFERENCES

1. Haffejee . Quarterly Journal of Medicine 1992. New series 84; 305: 641-658.
2. Ahmed Samir et al. Pediatric Annals 1992; 21: 835-842.
3. Spaun J et al. Bull Wld Hlth Org 1961; 24: 271-279.
4. The association of Clinical Pathologists 1961. Broadsheet 34.
5. Picard B et al. La Presse Medicale 1983; 23: 2-6.
6. Klein GC. Applied Microbiology 1971; 21: 999-1001.
7. Young DS. Effects of drugs on clinical laboratory test, 4th ed. AACC Press, 1995.

ATLAS Medical GmbH
Ludwig-Erhard Ring 3
15827 Blankenfelde-Mahlow
Germany
Tel: +49 - 33708 – 3550 30
Email: Info@atlas-medical.com
Website: www.atlas-medical.com

PPI2325A01
Rev A (05.01.2023)

REF	Catalogue Number		Temperature limit
IVD	*In Vitro* diagnostic medical device		Caution
	Contains sufficient for <n> tests and Relative size		Consult instructions for use (IFU)
LOT	Batch code		Manufacturer
	Fragile, handle with care		Use-by date
	Manufacturer fax number		Do not use if package is damaged
	Manufacturer telephone number		Date of Manufacture
	Keep away from sunlight		Keep dry
CONTROL+	Positive control	CONTROL-	Negative control

Atlas Medical

CRP LATEX KIT

IVD For *in -vitro* diagnostic and professional use only

2°C 8°C Store at 2-8°C. CE

INTENDED USE

CRP Latex kit is used to measure the CRP in human serum qualitatively and semi- quantitatively.

INTRODUCTION

C-reactive protein (CRP), the classic acute-phase of human serum, is synthesized by hepatocytes. Normally, it is present only in trace amounts in serum, but it can increase as much as 1,000-fold in response to injury or infection. The clinical measurement of CRP in serum therefore appears to be a valuable screening test for organic disease and a sensitive index of disease activity in inflammatory, infective and ischemic conditions. MacLeod and Avery found that antibody produced against purified CRP provided a more sensitive test than the C-polysaccharide assay. Since that time a number of immunological assays have been devised to measure CRP such as capillary precipitation, double immunodiffusion and radical immunodiffusion.

The CRP reagent kit is based on the principle of the latex agglutination assay described by Singer and Plotz. The major advantage of this method is the rapid two (2) minute reaction time.

PRINCIPLE

The CRP reagent kit is based on an immunological reaction between CRP Antisera bound to biologically inert latex particles and CRP in the test specimen. When serum CRP equal or greater than the Reagent sensitivity (Indicated on the label of the latex vial) the visible agglutination occurs.

MATERIALS

MATERIALS PROVIDED

- CRP Latex Reagent: Latex particles coated with goat IgG anti-human CRP (approximately 1 %), pH 8.2 MIX **WELL BEFORE USE.**
- CRP Positive Control Serum (Red Cap): A stabilized pre-diluted human serum containing >20mg/L CRP.
- CRP Negative Control Serum (Blue Cap): A stabilized pre-diluted animal serum.
- Glass Slides.
- Stirring Sticks.
- Package insert.

NOTE: This package insert is also used for individually packed reagent.

MATERIALS REQUIRED BUT NOT PROVIDED

- Mechanical rotator with adjustable speed at 80-100 r.p.m.
- Vortex mixer.
- Pippetes 50 µL.
- Glycine Buffer 20X (1000 mmol/L): add one part to nineteen parts of distilled water before use.

PACKAGING CONTENTS

REF **8.00.00.0.0100 (1x4ml Latex Reagent, 1x1ml positive control, 1x1ml negative control)**

PRECAUTIONS

- All reagents contain 0.1 %(w/v) sodium azide as a preservative.
- Protective clothing should be worn when handling the reagents.
- Wash hands and the test table top with water and soap once the testing is done.
- Reagents containing sodium azide may be combined with copper and lead plumbing to form highly explosive metal azides. Dispose of reagents by flushing with large amounts of water to prevent azide buildup.
- For In *Vitro* diagnostic use.
- Components prepared using human serum found negative for hepatitis B surface antigen (HBsAg), HCV and antibody to HIV (1/2) by FDA required test. However, handle controls as if potentially infectious.
- Accuracy of the test depends on the drop size of the latex reagent (40µl). Use only the dropper supplied with latex and hold it perpendicularly when dispensing.
- Use a clean pipette tip and stirring stick for each specimen, and glass slides should be thoroughly rinsed with water and wiped with lint-free tissue after each use.
- Check reactivity of the reagent using the controls provided.
- Do not use these reagents if the label is not available or damaged.
- Do not use the kit if damaged or the glass vials are broken or leaking and discard the contents immediately.
- Test materials and samples should be discarded properly in a biohazard container.

REAGENT PREPARATION:

The CRP Latex reagent is ready to use. No preparation is required. Mix gently before use to ensure a uniform suspension of particles.

STORAGE AND STABILITY

- Reagents are stable until specified expiry date on bottle label when stored refrigerated (2 - 8°C).
- **DO NOT FREEZE.**
- The CRP latex reagent, once shaken must be uniform without visible clumping. When stored refrigerated, a slight sedimentation may occur and should be considered normal.
- Do not use the latex reagent or controls if they become contaminated.
- Always keep vials in vertical position. If the position is changed, gently mix to dissolve aggregates that may be present.
- Reagents deterioration: Presence of particles and turbidity.

SPECIMEN COLLECTION AND STORAGE

- Use fresh serum collected by centrifuging clotted blood.
- If the test cannot be carried out on the same day, store the specimen for 7 days at 2-8°C and for 3 months at -20°C.
- Samples with presence of fibrin should be centrifuged before testing. Do not use highly hemolyzed or lipemic samples.
- Do not use plasma.

PROCEDURE

A. QUALITATIVE TEST:

1. Allow the reagents and samples to reach room temperature. The sensitivity of the test may be reduced at low temperatures.
2. Place (40 µL) of the sample and one drop of each Positive and Negative controls into separate circles on the slide test.
3. Mix the CRP-latex reagent vigorously or on a vortex mixer before using and add one drop (40 µL) next to the samples 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. Place the slide on a mechanical rotator at 80-100 r.p.m. for 2 minutes. False positive results could appear if the test is read later than two minutes.

B. SEMI-QUANTITATIVE TEST:

1. Make serial two-fold dilutions of the sample in 9 g/L saline solution.
2. Proceed for each dilution as in the qualitative method.

QUALITY CONTROL

- Positive and Negative controls are recommended to monitor the performance of the procedure, as well as a comparative pattern for a better result interpretation.
- All result different from the negative control result, will be considered as a positive.

READING AND INTERPRETATION

Examine macroscopically the presence or absence of visible agglutination immediately after removing the slide from the rotator.

The presence of agglutination indicates a CRP concentration equal or greater than the reagent sensitivity (mg/L CRP) (indicated on the label of the latex vial).

The titer, in semi-quantitative method, is defined as the highest dilution showing a positive result.

CALCULATIONS

The approximate CRP concentration in the patient sample is calculated as follows:

Sensitivity (Indicated on the label of the latex vial) x CRP Titer = mg/L

INTERFERENCES

NONE INTERFERING SUBSTANCES:

- Hemoglobin (10 g/dl)
- Bilirubin (20 mg/dl)
- Lipids (10 g/L)
- Other substances interfere, such as RF (100IU/ml).

NOTE

- High CRP concentration samples may give negative results. Retest the sample again using a drop of 20µl.
- The strength of agglutination is not indicative of the CRP concentration in the samples tested.
- Clinical diagnosis should not be made on findings of a single test result, but should integrate both clinical and laboratory data.

LIMITATIONS

1. Reaction time is critical. If reaction time exceeds two (2) minutes, drying of the reaction mixture may cause false positive results.
2. Freezing the CRP Latex Reagent will result in spontaneous agglutination.
3. Intensity of agglutination is not necessarily indicative of relative CRP concentration; therefore, screening reactions should not be graded.
4. A false negative can be attributed to a prozone phenomenon (antigen excess). It is recommended, therefore, to check all negative sera by retesting at a 1:10 dilution with glycine buffer.

.REFERENCE VALUES

Up to the reagent sensitivity (Indicated on the label of the latex vial). Each laboratory should establish its own reference range.

PERFORMANCE CHARACTERISTICS

- **Sensitivity:** Refer to vial label.
- **Prozone effect:** No prozone effect was detected up to 1600 mg/L
- **Diagnostic sensitivity:** 95.6 %.
- **Diagnostic specificity:** 96.2 %.

REFERENCES

1. Pepys, M.B.. Lancet 1:653 (1981).
2. Werner, M.. Clin.Chem. Acta 25:299 (1969).
3. MacLeod, C.M., et. al.. J. Exp. Med 73:191 (1941).
4. Wood, HF., et. al.. J. Clin. Invest. 30: 616 (1951).
5. Mancini, G., et. al. Immunochemistry 2:235 (1965).
6. Singer, J.M., et. al.. Am. J. Med 21: 888 (1956).
7. Fischer, C.L., Gill,. C.W.. In Serum Protein Abnormalities. Boston, Little, Brown and Co., (1975).

ATLAS Medical GmbH
Ludwig-Erhard Ring 3
15827 Blankenfelde-Mahlow
Germany
Tel: +49 - 33708 – 3550 30
Email: Info@atlas-medical.com
Website: www.atlas-medical.com

PPI2327A01
Rev A (05.01.2023)

REF	Catalogue Number		Temperature limit
IVD	*In Vitro* diagnostic medical device		Caution
Σ	Contains sufficient for <n> tests and Relative size		Consult instructions for use (IFU)
LOT	Batch code		Manufacturer
	Fragile, handle with care		Use-by date
	Manufacturer fax number		Do not use if package is damaged
	Manufacturer telephone number		Date of Manufacture
	Keep away from sunlight		Keep dry
CONTROL+	Positive control	CONTROL-	Negative control

Atlas Medical

RF LATEX KIT

IVD For In-*Vitro* diagnostic and professional use only

2°C 8°C Store at 2-8°C CE

INTENDED USE

Atlas RF latex test for the qualitative and semi-quantitative measurement of RF in human serum.

INTRODUCTION

Rheumatoid factors (RF) are antibodies directed against antigenic sites in the Fc fragment of human and animal IgG. Their frequent occurrence in rheumatoid arthritis makes them useful for diagnosis and monitoring of the disease.

One method used for rheumatoid factor detection is based on the ability of rheumatoid arthritis sera to agglutinate sensitized sheep red cells, as observed by Waaler and Rose. A more sensitive reagent consisting of biologically inert latex beads coated with human gamma globulin was later described by Singer and Plotz. The RF kit is based on the principle of the latex agglutination assay of Singer and Plotz. The major advantage of this method is rapid performance (2-minutes reaction time) and lack of heterophile antibody interference.

PRINCIPLE

The RF reagent is based on an immunological reaction between human IgG bound to biologically inert latex particles and rheumatoid factors in the test specimen. When serum containing rheumatoid factors is mixed with the latex reagent, visible agglutination occurs.

MATERIALS

MATERIALS PROVIDED

- RF Latex Reagent: Latex particles coated with human gamma-globulin, pH, 8,2. Preservative.
- RF Positive Control Serum (Red Cap): Human serum with a RF concentration > 30 IU/Ml. Preservative.
- RF Negative Control Serum (Blue Cap): Animal serum. Preservative.
- Glass Slide
- Stirring sticks

NOTE: This package insert is also used for individually packed reagent.

MATERIALS REQUIRED BUT NOT PROVIDED

- Mechanical rotator with adjustable speed at 80-100 r.p.m.
- Vortex mixer.
- Pippetes 50 µL
- Glycine Buffer 20x (1000mmol/L): add one part to nineteen parts of distilled water before use.

Packaging contents

REF **8.00.04.0.0100 (1x4ml Latex Reagent, 1x1ml positive control, 1x1ml negative control)**

PRECAUTIONS

- All reagents contain 0.1 %(w/v) sodium azide as a preservative.
- Protective clothing should be worn when handling the reagents.
- Wash hands and the test table top with water and soap once the testing is done.
- Reagents containing sodium azide may be combined with copper and lead plumbing to form highly explosive metal azides. Dispose of reagents by flushing with large amounts of water to prevent azide buildup.
- For In Vitro diagnostic use.
- Components prepared using human serum found negative for hepatitis B surface antigen (HBsAg), HCV and antibody to HIV (1/2) by FDA required test. However, handle controls as if potentially infectious.
- Accuracy of the test depends on the drop size of the latex reagent (40µl). Use only the dropper supplied with latex and hold it perpendicularly when dispensing.
- Use a clean pipette tip and stirring stick for each specimen, and glass slides should be thoroughly rinsed with water and wiped with lint-free tissue after each use.
- Check reactivity of the reagent using the controls provided.
- Do not use these reagents if the label is not available or damaged.
- Do not use the kit if damaged or the glass vials are broken or leaking and discard the contents immediately.
- Test materials and samples should be discarded properly in a biohazard container.

REAGENT PREPARATION:

- The RF Latex reagent is ready to use. No preparation is required. Mix gently before use to ensure a uniform suspension of particles.

STORAGE AND STABILITY

- Reagents are stable until specified expiry date on bottle label when stored refrigerated (2-8°C).
- Do not freeze.
- Always keep vials in vertical position. If the position is changed, gently mix to dissolve aggregates that may be present.
- The RF latex reagent, once shaken must be uniform without visible clumping. When stored refrigerated, a slight sedimentation may occur and should be considered normal.
- Do not use the latex reagent or controls if they become contaminated.
- Reagents deterioration: Presence of particles and turbidity.

SPECIMEN COLLECTION AND STORAGE

- Use fresh serum collected by centrifuging clotted blood.
- If the test cannot be carried out on the same day, store the specimen for 7 days at 2-8°C and for 3 months at -20°C.
- Samples with presence of fibrin should be centrifuged before testing. Do not use highly hemolyzed or lipemic samples.
- Do not use PLASMA.

PROCEDURE

Qualitative method

1. Allow the reagents and samples to reach room temperature. The sensitivity of the test may be reduced at low temperatures.
2. Place (40 µL) of the sample and one drop of each Positive and Negative controls into separate circles on the slide test.
3. Mix the RF-latex reagent rigorously or on a vortex mixer before using and add one drop (40 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. Place the slide on a mechanical rotator at 80-100 r.p.m. for 2 minutes. False positive results could appear if the test is read later than two minutes.

Semi-quantitative method

1. Make serial two-fold dilutions of the sample in 9 g/L saline solution.
2. Proceed for each dilution as in the qualitative method.

READING AND INTERPRETATION

Examine macroscopically the presence or absence of visible agglutination immediately after removing the slide from the rotator. The presence of agglutination indicates a RF concentration equal or greater than 8 IU/mL (Note 1).

The titer, in the semi-quantitative method, is defined as the highest dilution showing a positive result.

CALCULATIONS

The approximate RF concentration in the patient sample is calculated as follows:

8 x RF Titer = IU/mL

INTERFERENCES

NON-INTERFERING SUBSTANCES:

- Hemoglobin (10g/L)
- Bilirubin (20mg/dl)
- Lipids (10g/L)

Other substances may interfere.

QUALITY CONTROL

- Positive and Negative controls are recommended to monitor the performance of the procedure, as well as a comparative pattern for a better result interpretation.
- All result different from the negative control result, will be considered as a positive.

PERFORMANCE CHARACTERISTICS

Analytical sensitivity

8 (6-16) IU/ml, under the described assay conditions.

PROZONE EFFECT

No prozone effect was detected up to 1500 IU/ml.

DIAGNOSTIC SENSITIVITY

100%.

DIAGNOSTIC SPECIFICITY

100%.

The diagnostic sensitivity and specificity have been obtained using 139 samples compared with the same method of a competitor.

LIMITATIONS

- Reaction time is critical. If reaction time exceeds 2 minutes, drying of the reaction mixture may cause false positive result.
- Freezing the RF Latex Reagent will result in spontaneous agglutination.
- Intensity of agglutination is not necessarily indicative of relative RF concentration; therefore, screening reactions should not be graded.
- Increased levels of RF may be found in some diseases other than rheumatoid arthritis such as infectious mononucleosis, sarcoidosis, lupus erythematosus, Sjogren's syndrome.
- Certain patients with rheumatoid arthritis will not have the RF present in their serum.
- 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.
- Diagnosis should not be solely based on the results of latex method but also should be complemented with a Waaler Rose test along with the clinical examination.

REFERENCE VALUES

Up to 8 IU/mL. Each laboratory should establish its own reference range.

NOTES

1. Results obtained with a latex method do not compare with those obtained with Waaler Rose test. Differences in the results between methods do not reflect differences in the ability to detect rheumatoid factors.

REFERENCES

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. Robert H Shmerling et al. The American Journal of Medicine 1991; 91: 528 –534.
4. Adalbert F. Schubart et al. The New England Journal of Medicine 1959; 261: 363 – 368.
5. Charles M. Plotz 1956; American Journal of Medicine; 21:893 – 896.
6. Young DS. Effects of drugs on clinical laboratory test, 4th ed. AACC Press, 1995.

ATLAS Medical GmbH
Ludwig-Erhard Ring 3
15827 Blankenfelde-Mahlow
Germany
Tel: +49 - 33708 – 3550 30
Email: Info@atlas-medical.com
Website: www.atlas-medical.com

PPI2326A01
Rev A (05.01.2023)

REF	Catalogue Number		Temperature limit
IVD	*In Vitro* diagnostic medical device		Caution
	Contains sufficient for <n> tests and Relative size		Consult instructions for use (IFU)
LOT	Batch code		Manufacturer
	Fragile, handle with care		Use-by date
	Manufacturer fax number		Do not use if package is damaged
	Manufacturer telephone number		Date of Manufacture
	Keep away from sunlight		Keep dry
CONTROL+	Positive control	CONTROL-	Negative control