DRG® Antibody to Treponema Pallidum ELISA (EIA-4131)

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NAME AND INTENDED USE

Antibody to Treponema Pallidum (ELISA) is an *in vitro* enzyme immunoassay for the confirmatory detection of Anti-TP in human serum or plasma.

PRINCIPLE

The purified recombinant TP antigen is coated on the multi-wells. When serum sample and recombinant TP antigen labeled with HRP (conjugated) are added to the coated wells, and if TP antibody is present in the sample, a complex of TP Ag -Anti-TP-TP Ag labeled with HRP will form. This enzyme reaction produces a color change, and the intensity of the absorbance at 450nm(single wavelength) or 450/630nm(double wavelength) indicates the presence or absence of anti-TP in the sample. The test is special, sensitive, reproducible and easy to operate.

STORAGE AND STABILITY

Store the kit at 2~8°, protected from light. The kit is stable within the expiration date printed on kit boxes. Do not freeze or use the kit beyond the expiration date.

MATERIALS PROVIDED

1. Coated Microwell Plate (TP Antigen)	1 block (96 wells)
2. Enzyme Conjugant	1 bottle (10 ml)
3. Positive Control Serum	1 vial 0.5 ml)
4. Negative Control Serum	1 vial (0.5 ml)
5. Concentrated Wash Buffer (1:20 dilution prior to use)	1 bottle (30 ml)
6. Substrate A	1 bottle (6 ml)
7. Substrate B	1 bottle (6 ml)
8. Stop Solution	1 bottle (10 ml)
9. Seal Paper	1 piece

PRECAUTIONS

1. Bring Antibody to Treponema Pallidum (ELISA) (all reagents), and samples to room temperature before use (approximately 30 minutes), put the remained reagents to the sealed pouch, and return to 2~8° in time.
2. If the wash solution is not sufficient, prepare as following before use:
PH7.2, 0.1M PBS-0.5%Tween 20. Resolve the concentrated wash solution at 37° if crystals appear.
3. Do not interchange reagents between kit lots.
4. Handle all reagents, samples and controls as if capable of transmitting an infectious agent. It is recommended that these reagents and samples be handled using established good laboratory working practices.
5. The color change technique (Colorimetric Method) is used in the dilute solution for sample. After original serum or plasma sample is added, the color turns from yellowish green to blue purple. If the serum or plasma is non-original, it is normal that there is little change or other color.

6. The seal paper can't be used repeatedly.
7. Dilute the wash solution with distilled water to 1: 20 prior to use.
8. Do not use the kit beyond its expiration date. The date is printed on kit boxes.
9. The NaN_3 can't be used to preserve the reagents.
10. The shelf life is 12 months.

TEST PROCEDURE

1. Bring all reagents and samples to room temperature approximately 30 minutes before use, dilute the concentrated wash solution with distilled water to 1: 20
2. For each test, set one blank, two positive and two negative controls. Add 50µl positive and negative control serum into positive and negative control wells respectively, add 50µl test serum into test wells, add 50µl enzyme conjugant into test wells (The blank well is omitted), mix thoroughly, incubate for 30 minutes at 37°.
3. Discard the liquid in all wells and bring them to dry. Fill the wells with wash solution (>300µl per well), and do not let it spill out. Lay aside for 5-10 seconds, discard the liquid in all wells and bring them to dry. Repeat 5 times.
4. Add substrate A 50µl to each well, then add substrate B 50µl to each well, mix thoroughly, and incubate for 15 minutes at 37° protected from light.
5. Add 50µl stop solution to each well, mix thoroughly.
6. Measure the absorbance at 450nm (single wavelength) against the blank or 450/630nm (double wavelength).

INTERPRETATION OF RESULTS

Colorimetric Method

Cut Off Value calculation:

COV = the average OD of negative controls + 0.10

Positive OD_{450} of sample ≥ COV

Negative OD_{450} of sample < COV

Invalid If the average OD of positive controls is below or equal to 0.80, the result is invalid. In any event, repeat the test. If the problem persists, contact the local distributor.

According to the test design, the margin between OD of positive controls and negative controls should be above 0.80; otherwise, the result of this test is fallacious.

Notes If the absorbance of negative controls is below 0.05, calculate it as 0.05. If the absorbance of negative controls is above 0.05, calculate it as its original value.

PERFORMANCE CHARACTERISTICS

Sensitivity the agreement rate of the tests to 10 positive anti-TP reference serum ≥10/10 (100%)

Specificity the agreement rate of the tests to 20 negative anti-TP reference serum ≥20/20 (100%)

Precision CV(%)≤15% (n=10)