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User's Manual

Human Schistosoma japonicum-Ag ELISA Kit

REF **DEIASL190**

Σ **96T**

RUO

This product is for research use only and is not intended for diagnostic use.

For illustrative purposes only. To perform the assay the instructions for use provided with the kit have to be used.

Creative Diagnostics

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PRODUCT INFORMATION

Intended Use

This Schistosoma japonicum-Ag ELISA kit is intended for qulitative detection of Schistosoma japonicum in human serum and plasma.

General Description

A genus of trematodes, Schistosoma, commonly known as blood-flukes and bilharzia, includes flatworms which are responsible for a highly significant parasitic infection of humans by causing the disease schistosomiasis, and is considered by the World Health Organization as the second most s°Cioeconomically devastating parasitic disease, next only to malaria, with hundreds of millions infected worldwide. Adult worms parasitize mesenteric blood vessels. They are unique among trematodes or any other flatworms in that they are dioecious with distinct sexual dimorphism between male and female. Eggs are passed through urine or feces to fresh water, where larva must pass through an intermediate snail host, before a different larval stage of the parasite emerges that can infect a new mammalian host by directly penetrating the skin.

Principles of Testing

The Stop Solution changes the color from blue to yellow and the intensity of the color is measured at 450 nm using a spectrophotometer. In order to measure the concentration of Schistosoma japonicum-Ag in the sample, this Schistosoma japonicum-Ag ELISA Kit includes a set of calibration standards. The calibration standards are assayed at the same time as the samples and allow the operator to produce a cutoff value. The existence or not of Schistosoma japonicum-Ag in the samples is then determined by comparing the O.D. of the samples to the CUT OFF.

Reagents And Materials Provided

1. Microelisa stripplate: 12*8strips
2. Negative control: 0.5ml
3. Positive control: 0.5ml
4. HRP-Conjugate reagent: 10.0ml
5. 20X Wash solution: 25ml
6. Sample Diluent: 6.0ml
7. Chromogen Solution A: 6.0ml
8. Chromogen Solution B: 6.0ml
9. Stop Solution: 6.0ml
10. Closure plate membrane
11. User manual
12. Sealed bags

Materials Required But Not Supplied

1. Standard microplate reader(450nm)
2. Precision pipettes and Disposable pipette tips.
3. 37°C incubator

Storage

Store at 4°C for frequent use, at -20°C for infrequent use. Avoid multiple freeze-thaw cycles.

Specimen Collection And Preparation

Serum - Use a serum separator tube and allow samples to clot for 30 minutes before centrifugation for 10 minutes at approximately 3000×g. Remove serum and assay immediately or aliquot and store samples at -20°C or -80°C. Avoid repeated freeze-thaw cycles.

Plasma - Collect plasma using EDTA or heparin as an anticoagulant. Centrifuge samples for 30 minutes at 3000×g at 2-8°C within 30 minutes of collection. Store samples at -20°C or -80°C. Avoid repeated freeze-thaw cycles.

Cell culture supernates and other biological fluids - Remove particulates by centrifugation and assay immediately or aliquot and store samples at -20°C or -80°C. Avoid repeated freeze-thaw cycles.

Note: The samples should be centrifugated dequately and no hemolysis or granule was allowed.

Reagent Preparation

20×wash solution: Dilute with Distilled or deionized water 1:20.

Assay Procedure

1. Prepare all reagents before starting assay procedure. It is recommended that all Standards and Samples be added in duplicate to the Microelisa Stripplate.
2. Separately add Positive control and Negative control 50µl to the Positive and Negative well; Add testing sample 10µl then add Sample Diluent 40µl to testing sample well.
3. Add 100µl of HRP-conjugate reagent to each well, cover with an adhesive strip and incubate for 60 minutes at 37°C.
4. Aspirate each well and wash, repeating the process four times for a total of five washes. Wash by filling each well with Wash Solution (400µl) using a squirt bottle, manifold dispenser or autowasher. Complete removal of liquid at each step is essential to good performance. After the last wash, remove any remaining Wash Solution by aspirating or decanting. Invert the plate and blot it against clean paper towels.
5. Add chromogen solution A 50µl and chromogen solution B 50µl to each well. Gently mix and incubate for 15 minutes at 37°C. Protect from light.
6. Add 50µl Stop Solution to each well. The color in the wells should change from blue to yellow. If the color in the wells is green or the color change does not appear uniform, gently tap the plate to ensure thorough

mixing.

7. Read the Optical Density (O.D.) at 450 nm using a microtiter plate reader within 15 minutes.

Quality Control

Test validity: the average of Positive control well≥1.00; the average of Negative control well ≤0.15.

Interpretation Of Results

Calculate Critical (CUT OFF): Critical= the average of Negative control well + 0.15.

Negative Result: sample OD< Calculate Critical (CUT OFF) is Negative.

Positive Result: sample OD≥ Calculate Critical (CUT OFF) is Positive.

Precautions

1. Do not substitute reagents from one kit to another. Standard, conjugate and microplates are matched for optimal performance. Use only the reagents supplied by manufacturer.
2. Do not remove microplate from the storage bag until needed. Unused strips should be stored at 2-8°C in their pouch with the desiccant provided.
3. Mix all reagents before using. Remove all kit reagents from refrigerator and allow them to reach room temperature (20-25°C).