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Human Tetanus IgG Quantitative Elisa Kit

Product Number: TQE01-96T

Shipping and Storage

1. 2-8℃, stored away from light and moisture
2. Validity period: 6 months

Components

Components	96 T	48T
Microelisa stripplate	12*8strips	12*4strips
Negative control	0.5ml	0.5ml
Positive control	0.5ml	0.5ml
Detect antibody HRP	10mL	5mL
20×Buffer WB	25mL	15mL
Chromogen Solution A	6mL	3mL
Chromogen Solution B	6mL	3mL
Stop Solution	6mL	3mL
Closure plate membrane	2pcs	2pcs

Note:20×Buffer WB:Dilute distilled water at a ratio of 1:20, which is 1 part of 20×Buffer WB and 19 parts of distilled water.

Description

The reagent kit adopts a double antibody one-step sandwich enzyme-linked immunosorbent assay (ELISA). Add the sample and HRP labeled detection antibody to the pre coated micro well with tetanus antigen, incubate and thoroughly wash. Using substrate TMB for color development, TMB is converted to blue under the catalysis of peroxidase and to the final yellow under the action of acid. Measure the absorbance (OD value) at a wavelength of 450nm using an enzyme-linked immunosorbent assay (ELISA) reader, and compare it with the CUTOFF value to determine the presence or absence of tetanus IgG antibody in the specimen.

Sample collection and storages

1. **Serum:** Use test tubes free of pyrogens and endotoxins, avoid any cell irritation during the operation, collect blood, centrifuge at 3000 rpm for 10 minutes, and quickly and carefully separate serum and red blood cells.
2. **Plasma:** EDTA、Citrate or heparin anticoagulant. Centrifuge at 3000 rpm for 30 minutes and collect the supernatant.
3. **Cell supernatant:** Centrifuge at 3000 rpm for 10 minutes to remove particles and polymers.
4. **Tissue homogenat**e: Crush the tissue by adding an appropriate amount of physiological saline. Centrifuge at 3000 rpm for 10 minutes and collect the supernatant.
5. **Storage:** If the sample is not tested in a timely manner after collection, please divide it into batches according to a single dose, freeze it at -20 ℃, avoid repeated freezing and thawing, thaw it at room temperature, and ensure that the sample is evenly filled and thawed.

Materials required but not supplied

1. ELISA reader (450nm)
2. High precision sampler and nozzle: 0.5-10uL, 2-20uL, 20-200uL, 200-1000uL
3. 37 ℃ constant temperature box

Note

For Research Use Only

1. The reagent kit should be stored at 2-8 ℃ and equilibrated at room temperature for 20 minutes before use. The concentrated washing solution taken out of the refrigerator will have crystals, which is a normal phenomenon. Heating in a water bath will completely dissolve the crystals before use.
2. The Flat noodles not used in the experiment shall be immediately put back into the self sealing bag and sealed (low-temperature dry) for storage.
3. The preprocessed sample does not need to be diluted, just add 50μL directly.
4. Strictly follow the time, liquid dosage, and sequence indicated in the instructions for incubation operation.
5. Shake all liquid components thoroughly before use.

Washing method

1. Manual board washing: Shake off the liquid in the holes, fill each hole with Buffer WB, let it stand for 1 minute, shake off the liquid in the holes, pat dry on absorbent paper, and wash the board 5 times in this way.
2. Automatic washing machine: Inject 350μL of Buffer WB into each well, soak for 1 minute, and wash the plate 5 times.

Protocol

1. Take out the Microelisa stripplate from the aluminum foil bag after 20 min of room temperature balance, and seal the remaining Flat noodles with a self sealing bag and put it back at 4 ℃.
2. Set up negative and positive control wells and sample wells, and add 50μL of negative control and 50μL of positive control to each well;
3. Add 50μL of the test sample to the test well;
4. Subsequently, 100μL of horseradish peroxidase (HRP) labeled detection antibody was added to each well of the negative, positive control, and sample wells. The reaction wells were sealed with a sealing membrane and incubated at 37 ℃ in a water bath or constant temperature incubator for 60 minutes.
5. Discard the liquid, pat dry on absorbent paper, fill each well with Buffer WB, let it stand for 1 minute, shake off Buffer WB, pat dry on absorbent paper, repeat washing the board 5 times (or use a board washing machine).
6. Add 50μL of substrate A and B to each well, and incubate at 37 ℃ in the dark for 15 minutes.
7. Add 50μL of termination solution to each well and measure the OD value of each well at a wavelength of 450nm within 15 minutes.

Determine the result

1. **Experimental effectiveness:** The average OD value of the positive control wells is ≥ 1.00;
2. The average OD value of negative control wells is ≤ 0.15.
3. **Cut off calculation:** Cut off=Average value of negative control wells+0.15
4. **Negative judgment:** If the OD value of the sample is less than the critical value (Cut off), the sample is negative
5. **Positive judgment:** If the sample OD value is greater than the critical value (Cut off), the sample is positive

Performance of kit

1. **Accuracy:** The average OD value of positive control wells is ≥ 1.00; The average OD value of the negative control wells is ≤ 0.15, indicating the validity of the test results.
2. **Specificity:** Does not cross react with other soluble structural analogues.
3. **Repeatability:** The coefficient of variation within and between plates is less than 15%.