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Human Hantavirus (HV) ELISA Kit (Qualitative)

Product Number: ELK12J0991

Shipping and Storage

1. The components of the reagent kit should be stored at 2-8°C;
2. Before use, please ensure that the reagent kit is stored according to the specified conditions, and only prepare the required amount of reagents for each experiment;
3. Preparation of 1×Wash Solution: If crystals precipitate from the concentrated wash solution (20×), it needs to be heated at 37°C until all crystals are dissolved before use. Dilute with distilled water at a ratio of 1:20, for example: add 10mL of concentrated washing solution to 190mL of distilled water and mix evenly. The prepared washing solution can be stored at room temperature for one week.

Component

Component	Kit Components	48T	96T
Microplate stripplate	Wrapping board	8×6 strips	8×12 strips
Positive control	Positive control	0.5mL	1mL
Negative control	Negative control	0.5mL	1mL
Enzyme markers	HRP enzyme labeled anti human IgG	5mL	10mL
Sample diluent	PBS, BSA, etc	5mL	10mL
20×washing buffer	20×washing buffer	15mL	25mL
Chromogenic agent	H_2O_2, TMB	6mL	10mL
Stop Solution	Dilute sulfuric acid, anti precipitation agent	3mL	6mL

Note: If you need to purchase universal components separately, please provide the corresponding component names.

Description

The kit employs the indirect method principle: Hantaan virus recombinant antigens are coated on the enzyme immunoassay plate. Samples are added, and IgG antibodies in the samples are captured to form "antigen-IgG antibody" immune complexes. Free impurities are washed away, followed by the addition of HRP-labeled enzyme-conjugated antibodies to create "antigen-IgG antibody-enzyme-conjugated antibody" sandwich complexes. Excess enzyme-conjugated antibodies are then washed away. TMB is added for color development, and a stop solution is subsequently added. The OD value is measured at 450nm using a microplate reader. The intensity of the color is directly proportional to the concentration of the target antibody in the sample.

Application

This kit is designed for the detection of human hantavirus IgG antibodies in serum and plasma, for research purposes only, not for diagnostic use.

Specimen collection and storage

The sample collection and storage conditions listed below are intended as general guidance, and the stability of storage and use of different types of samples has not been fully evaluated.

1. Serum: Collect whole blood using a serum collection tube, let it stand at room temperature for blood cell agglutination, or centrifuge directly at 1000×g for 15 minutes to obtain serum. The operation should be gentle to avoid hemolysis. The obtained serum should be tested in a timely manner. If it cannot be tested in a timely manner, it should be divided into multiple parts and stored at a temperature of -20°C or below for no more than 6 months. The frozen storage of serum samples should not exceed 2 freeze-thaw cycles.
2. Plasma: Collect plasma using EDTA or heparin blood collection tubes. After collecting whole blood, centrifuge at 1000×g for

15 minutes to obtain plasma. The operation should be gentle to avoid hemolysis. The obtained plasma should be tested in a timely manner. If it cannot be tested in a timely manner, it should be divided into multiple parts and stored at a temperature of -20°C or below for no more than 6 months. The frozen plasma samples should not be thawed more than twice.

3. Cannot detect samples containing sodium azide, suspended fibrin or aggregates, and severe hemolysis.
4. The sample should be free from microbial contamination.

Materials required but not supplied

1. Distilled water or deionized water, disposable centrifuge tubes, disposable gloves and other consumables;
2. Pipettes, multi-channel pipettes, and matching gun heads;
3. Beakers, measuring cylinders, reagent bottles and other containers;
4. Sealing film or other alternative materials used for sealing microporous plates;
5. Microplate oscillator (if needed), centrifuge, vortex oscillator and other auxiliary equipment;
6. Constant temperature incubator, constant temperature water bath, enzyme-linked immunosorbent assay reader, washing machine.

Protocol

Before use, allow all reagents to equilibrate at room temperature for about 30 minutes, or quickly return to room temperature in a 37°C incubator.

Note: Do not open the enzyme-linked immunosorbent assay (ELISA) plate until it has returned to room temperature.

1. Reasonably arrange positive control wells, negative control wells, blank wells, and sample wells.
2. Add 100μL of positive control to the positive control well and 100uL of negative control to the negative control well. Set 2 wells for each positive and negative control well.
3. Add 100μL of sample diluent to the sample well first, and then add 10μL of sample; Blank wells without samples or enzymes, other operations are the same as sample wells.
4. Cover with a sealing film and incubate at 37°C for 30 minutes.
5. Wash the plate 5 times, discard the residual washing solution after the last washing, invert the enzyme-linked immunosorbent assay plate, and pat dry on clean absorbent paper.
6. Add 100μL of enzyme labeled working solution to each well.
7. Cover with a sealing film and incubate at 37°C for 30 minutes.
8. Wash the plate 5 times, discard the residual washing solution after the last washing, invert the enzyme-linked immunosorbent assay plate, and pat dry on clean absorbent paper.
9. Add 100μL of color developer to each well
10. Incubate at 37°C in the dark for 15 minutes.
11. Add 50μL of termination solution to each well.
12. Single wavelength detection: Use an enzyme-linked immunosorbent assay (ELISA) reader to measure the absorbance value at a wavelength of 450nm; Dual wavelength detection: Use an enzyme-linked immunosorbent assay (ELISA) reader to measure absorbance values at 450/630nm wavelengths.

Positive judgment value Cut Off

Cut Off=Mean OD value of negative control wells multiplied by 2.1+0.1 (when the mean OD value of negative control wells is less than 0.050, it is calculated as 0.050).

Example: If the average OD value of the negative control well is 0.100, then the Cut OFF value is 0.100 × 2.1+0.1=0.310;

The average OD value of the negative control well is 0.020, so the Cut OFF value is 0.050 × 2.1+0.1=0.205.

Result judgment

1. Experimental conditions: Positive control OD value>1.0, and negative control OD value<0.15;

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2. Positive result: If the sample OD value is greater than the Cut OFF value, the result is considered positive;
3. Negative result: If the sample OD value is ≤ Cut OFF value, the result is considered negative.

Limitations of testing

The sample concentration is below the detection limit, and the detection results cannot be accurately judged. This reagent is a qualitative reagent and cannot be used as a quantitative reagent.

Test kit performance

1. The following results are based on the concentration values of the calibration standard and do not take into account the potential effects of other factors such as matrix effects.
2. Three positive reference products from enterprises were tested, and the results were all positive. Three negative reference products from the enterprise were tested, and the results were all negative.
3. Precision: The intra board precision CV is less than 10% (N=20), and the inter board precision CV is less than 15% (N=20).
4. Analytical specificity: Other related factors were prepared in dilution buffer at a concentration of 10μg/mL and no significant cross reactivity was observed.

Note

1. All ingredients in the reagent kit are for research purposes only!
2. The termination solution contains dilute sulfuric acid, which is corrosive and should be handled with caution.
3. The reagent kit contains preservatives, protein components, etc., which may cause skin allergic reactions. Masks should be worn to avoid inhaling light mist.
4. Color developing agents and concentrated washing solutions may cause skin, eye, and respiratory irritation. Masks should be worn to avoid inhaling light mist.
5. Wear protective gloves, protective clothing, eye and facial protection equipment, and wash hands thoroughly after experimental operations.

Disclaimers

1. Different batches of kits cannot be mixed. It is not recommended to assemble microporous Flat noodles on different plates into one plate for detection. Although the reagents of the same batch are used, there may be differences between plates.
2. During each experiment, a standard curve should always be prepared for parallel testing, and the previous standard curve should not be used for calculating the results of the next experiment.
3. Do not use expired reagent kits for testing.
4. The substrate color reagent should be colorless. If it turns blue, it indicates that it has deteriorated and cannot be used in experiments.
5. Use appropriate sealing membrane to seal microporous Flat noodles, so that the test results are more reliable.
6. Try to avoid generating bubbles during operation. Do not cross use the caps of different reagent bottles.
7. Experiments should be conducted in accordance with the instructions provided. Failure to follow the instructions may result in changes to the experimental results. Any experimental operations that do not follow the instructions should seek advice from after-sales technical support in advance. Otherwise, we cannot guarantee the reliability of the experimental results.