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Enterovirus Type 71 (EV71) Quantitation Kit (ELISA)

【Generic Name】

Enterovirus Type 71 (EV71) Quantitation Kit

Cat#: 17-0008

【Intended Use】

To detect the contents of EV71 in samples.

【Principle】

The EV71 Quantitation Kit is designed for the sensitive and specific detection of EV71 in an antibody sandwich format. Briefly, flat-bottom 96-well plates are coated with Anti-EV71 monoclonal antibody (mAb) that captures EV71 in the sample. A quick wash removes any unbound virus particles. Captured EV71 is detected by a second specific Anti-EV71 mAb, which is conjugated to horseradish peroxidase (HRP). Finally, the chromogenic substrate 3,3',5,5'-tetramethylbenzidine (TMB) is added. The amount of EV71 is proportional to the color generated in the coupled oxidation-reduction reaction and can be determined using a standard curve generated with known amounts of EV71.

【Materials and Reagents】

1. Coated plate, 12 wells × 8 strips
2. Anti-EV71 Detection mAb, HRP Conjugate, 12ml×1 vial
3. 20× Washing Buffer, 50mL×1 vial
4. Sample Dilution Buffer, 12ml×1 vial
5. Substrate Solution A, 7mL×1 vial
6. Substrate Solution B, 7mL×1 vial
7. Termination Solution, 7mL×1 vial
8. Sealing plate film, 2 pieces
9. Instruction book

【Storage】

1. All components remain stable for six months under the temperature condition 2-8°C;
2. Return each component to 4°C immediately after use

【Protocol for Detection】

1. Before using, pre-warm all the reagents to room temperature (18~25°C). Dilute 20× Washing Buffer to 1000ml and mix well.
2. Dilute samples with Sample Dilution Buffer and add 100 μl into each well of the ELISA plate. Set negative control and incubate at 37°C for 60min.
3. Remove samples from wells and wash all wells five times with prepared 1× Washing Buffer. Remove residual solution with paper pat.
4. Add 100 μl of HRP-Conjugated Anti-EV71 Detection mAb to each well. (Careful not to touch or scratch the surface of the wells). Incubate plate at 37°C for 45min.
5. Remove samples from wells and wash all wells five times with prepared 1× Washing Buffer. Remove residual solution with paper pat.
6. Add 50 μl each of Substrate Solution A and Substrate Solution B into each well. Mix thoroughly with shaking. Incubate at 37°C for 10~15min. Avoid light.
7. Stop the reaction by adding 50μl of Termination Solution.
8. Record the absorbance at 450nm on a plate reader within 30min.

【Result estimate】

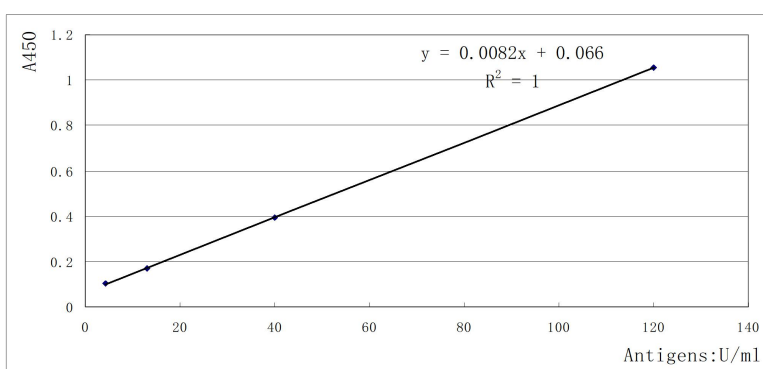
Cut off value=2.1 X N (N is A450 for negative control. set N=0.05, if N<0.05)

If A450 for negative control is larger than 0.15, this assay is failed.

If sample A450 is smaller than cut off value, this sample is negative. And vice versa.

Sensitivity: 12U/ml

Liner Range: 4~120U/ml



【Limitations】

1. This kit is only used to detect the content of EV71 in samples.
2. Results out of the measurement range of the kit are unreliable.
3. Severe hemolysis, chyle, and bilirubin samples may cause abnormal test results.
4. This kit is developed for in vitro research only

【Caution】

1. Avoid cross contamination.
2. Follow reader measure as a standard.
3. All samples and buffers should be added or removed with pipette.
4. Do not mix reagents from different batches.