



# SZABO SCANDIC

Part of Europa Biosite

## Produktinformation



Forschungsprodukte & Biochemikalien



Zellkultur & Verbrauchsmaterial



Diagnostik & molekulare Diagnostik



Laborgeräte & Service

Weitere Information auf den folgenden Seiten!  
See the following pages for more information!



### Lieferung & Zahlungsart

siehe unsere [Liefer- und Versandbedingungen](#)

### Zuschläge

- Mindermengenzuschlag
- Trockeneiszuschlag
- Gefahrgutzuschlag
- Expressversand

### SZABO-SCANDIC HandelsgmbH

Quellenstraße 110, A-1100 Wien

T. +43(0)1 489 3961-0

F. +43(0)1 489 3961-7

[mail@szabo-scandic.com](mailto:mail@szabo-scandic.com)

[www.szabo-scandic.com](http://www.szabo-scandic.com)

[linkedin.com/company/szaboscandic](https://www.linkedin.com/company/szaboscandic) 

**(FOR RESEARCH USE ONLY. DO NOT USE IT IN CLINICAL DIAGNOSIS !)**

## **Human Enterovirus 71 (EV71) IgM ELISA Kit**

Catalog No: E-HD-E129

96T/96T\*2

This manual must be read attentively and completely before using this product.

If you have any problems, please contact our Technical Service Center for help.

Toll-free: 1-888-852-8623 Tel: 1-832-243-6086 Fax: 1-832-243-6017

Email: [techsupport@elabscience.com](mailto:techsupport@elabscience.com)

Website: [www.vetassay-elab.com](http://www.vetassay-elab.com)

Please kindly provide us the lot number (on the outside of the box) of the kit for more efficient service.

**Test principle**

This ELISA kit uses Capture-ELISA as the method to detect the EV71-IgM in the sample. The micro-plate is pre-coated with Mouse-anti-human IgM ( $\mu$  chain). EV71-IgM in samples will be captured after adding the samples to wells. After washing completely, add EV71 antigen conjugated Horseradish Peroxidase (HRP), and it will react with the “anti- $\mu$  chain-EV71 IgM” compound to form “anti- $\mu$  chain-EV71 IgM-HRP conjugated EV71 antigen”. Wash and remove the free components. Add the TMB substrate to initiate color developing reaction. The presence of EV71-IgM antibody can be determined according to the OD value after colorimetric assay with the Micro-plate Reader.

**Kit components**

Item	Specifications
ELISA Micro-plate	96 wells
HRP Conjugated Working Solution	12 mL
20×Concentrated Wash Buffer	50 mL
Sample Diluent	12 mL
Substrate Reagent A	6 mL
Substrate Reagent B	6 mL
Stop Solution	6 mL
Positive Control	1 mL
Negative Control	1 mL
Plate Sealer	3 pieces
Sealed Bag	1 copy
Manual	1

**Experimental instrument**

Micro-plate Reader with 450 nm wavelength filter or dual-wavelength (450/630 nm)

High-precision transferpettor, EP tubes and disposable pipette tips

37°C Incubator or water bath

Deionized water

Absorbent paper

Loading slot for Wash Buffer

## Sample preparation

1. Fresh collected serum specimens should be fully centrifuged, then take clear liquid for test, if not fully precipitated, suspended fibrous protein may cause a false positive.
2. Anticoagulants (heparin, EDTA, sodium citrate) has no interference to the test results. In general, endogenous interferent (such as blood lipids, cholerythrin, hemoglobin) in sample, RF positive factors, pregnant sample and AFP positive sample, the antibody positive sample of related diseases (HAV, HCV, hepatitis B, syphilis, rubella, VZV, etc.) will not interfere the detection result.
3. Do not use heat-inactivated samples, heat inactivation will lead the degradation of IgM in sample.
4. Samples can be stored at 2-8°C for one week. If samples not tested in a week, store them at below -15°C and avoid freeze-thaw cycles.

## Assay procedure

Bring all reagents to room temperature for 30 min. Dilute the 20×Concentrated Wash Buffer for 20 times with distilled water.

### 1. Add sample:

- (1) Take out Micro-plate and mark it, reserve 1 well for blank control (empty), 3 wells for negative control, 1 well for positive control (100 µL control serum). (Blank well is not necessary for dual-wavelength detection)
- (2) Dilute the tested **Serum** with **Sample Diluent** at 1:10 into sample well (add 100 µL of sample diluent and add 10 µL of sample), mix fully.
- (3) Gently tap the plate to ensure thorough mixing.

**2. Incubate:** Cover the ELISA plate with sealer. Incubate for 30 min at 37°C.

**3. Wash:** After incubation, remove the plate sealer and aspirate the liquid of each well. Repeat the washing procedure for 5 times with wash buffer and immerse for 30-60 sec each time.

**4. HRP conjugate:** Add 100 µL of HRP Conjugate working solution to each well except the blank control well.

**5. Incubate:** Cover the ELISA plate with sealer. Incubate for 30 min at 37°C.

**6. Wash:** After incubation, remove the plate sealer and aspirate the liquid of each well. Repeat the washing procedure for 5 times with wash buffer and immerse for 30-60 sec each time.

**7. Add substrate:** Add 50 µL of Substrate Reagent A and 50 µL of Substrate Reagent B to each well. Gently tap the plate to ensure thorough mixing. Cover with a new plate sealer. Incubate for 15 min at 37°C in dark.

**8. Stop reaction:** Add 50 µL of Stop Solution to each well, gently tap the plate to ensure thorough mixing.

**9. OD Measurement:** Set the Micro-plate Reader wavelength at 450 nm (it is recommended to set the dual wavelength at 450 nm/630 nm) to detect A value of each well. Blank well is not needed when using dual wavelength 450 nm/630 nm for detection.

## Reference value

### 1. Result analysis

- (1) Use each test result independently. Determine the result according to the Cut Off value.
- (2) Calculate the Cut Off:  $\text{Cut Off (C.O)} = 0.10 + \text{negative control (NC) average A value}$  (when NC average  $A_{450} < 0.05$ , calculate at 0.05; while NC average  $A_{450} \geq 0.05$ , calculate at the actual value).

### 2. Quality control

- (1) Blank well (just chromogenic agent and stop solution) absorbance  $\leq 0.08$ .
- (2) Positive control (PC)  $A_{450} > 0.30$ .
- (3) Negative control (NC)  $A_{450} < 0.10$ .

The experimental result is valid if quality control is valid.

### 3. Determination of results

- (1) Positive result: Sample absorbance  $\geq$  Cut Off.
- (2) Negative result: Sample absorbance  $<$  Cut Off.

## Interpretation of test results

1. Negative result indicates no EV71-IgM antibody detected in samples, while positive result is just the opposite.
2. The concentration of EV71-IgM may be very low during the early stage of infection, it may cause negative result. The patient should recheck the EV71-IgM level in 7~14 days.
3. The reference value of the serological antibody testing for patients with immunity damage or accepted immunosuppressive therapy is limited.
4. The positive result of EV71-IgM may appear during the primary infection, as well as previous infection.

## Limitations of test method

1. This test is only used as the qualitative detection of EV71-IgM antibodies in serum of human.
2. The detection results of this kit are only for reference. For confirmation of the result, please combine the symptoms and other methods of detection, this detection cannot be used as the only criteria for result.

## Notes

1. Please read the manual carefully before use, changes of operation may result in unreliable results.
2. Wear gloves and work clothes during experiment, and the disinfection and isolation system should be strictly executed. All the waste should be handled as contaminant.
3. The stop solution is corrosive, it should be avoided to contact with skin and clothing. Wash immediately with plenty of water if contact it carelessly.
4. The ELISA Microtiter plate obtained from cold storage conditions should be adjusted to room temperature before use. The unused plate should be kept in a sealed bag with desiccant.
5. Concentrated washing liquid at low temperature condition is easy to crystallization, it should be adjusted to room temperature in order to dissolve completely before use.
6. The results shall depend on the readings of the micro-plate Reader.
7. **Each reagent is optimized for use in the E-HD-E129. Do not substitute reagents from any other manufacturer into the test kit. Do not combine reagents from other E-HD-E129 with different lot numbers.**
8. All the samples and waste material should be treated as infective material according to the relevant rules of biosafety.

## Storage and expiry date

Store unopened at 2 to 8°C. Do not freeze.

Please store the opened plate at 2~8°C, the shelf life of the opened kit is up to 1 month.

**Expiry date:** expiration date is on the box.