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**Diagnosis for chikungunya fever  
(English Translation)**

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### Foreword

This standard was drafted in accordance with the rules given in GB/T 1.1-2009.

This standard was drafted. Guangdong Provincial Center for Disease Control and Prevention, Guangzhou Eighth People's Hospital, China Center for Disease Control and Prevention, China Center for Disease Control and Prevention, Center for Disease Control and Prevention, Dongguan People's Hospital.

### 1 Scope

This standard specifies the diagnostic basis, diagnostic principles, diagnosis and differential diagnosis of Chikungunya fever.

This standard is applicable to the diagnosis of Chikungunya fever by various medical and health institutions at all levels and their medical staff.

### 2 Abbreviations

The following abbreviations apply to this document.

CHIKV. chikungunya virus ELISA. enzyme-linked immunosorbent assay IgG. immunoglobulin G (immunoglobulin G) IgM. immunoglobulin M (immunoglobulin M) RNA.

ribonucleic acid RT-PCR. reverse transcription-polymerase chain reaction

### **3.1 Epidemiological history**

Within 12 days before the onset of the disease, the disease occurred in the epidemic area of Chikungunya or around the workplace or workplace.

### **3.2 Clinical manifestations**

#### **3.2.1**

Fever. Arouse high fever, body temperature can reach above 39 °C, generally fever 1 d ~ 7 d. Some patients re-emerged after fever, table it is now a bimodal fever and returns to normal for 3 to 5 days. Often accompanied by chills, headache, back pain, muscle pain, photophobia, nausea, vomiting and other symptoms.

#### **3.2.2**

Joint pain. joint pain mainly involves small joints such as wrists and toes, and may also involve large joints such as knees and shoulders.

Severe pain is an important feature of this disease. Pain or arthritis in multiple joints in the acute phase, may be swollen or stiff, and heavier in the morning.

Severe cases can not be active, usually relieved from 1 week to 3 weeks. In some cases, joint pain can last for several months.

#### **3.2.3**

Rash. 2 to 5 days after onset, more than half of the cases have red maculopapular rash or purpura on the trunk, extremities, palms and soles.

The skin between the rash is mostly normal, and some of it is accompanied by itching sensation. It disappears after a few days and may be accompanied by scaling.

### **3.3 Laboratory inspection**

#### **3.3.1**

Positive for serum-specific IgM antibodies in the acute phase (see A.1 in Appendix A).

#### **3.3.2**

The titer of serum-specific antibody (IgG or neutralizing antibody) during the recovery period is 4 times higher than the acute phase, or the antibody is negative in the acute phase.

The antibody in the recovery phase is positive (see A.2, A.3 in Appendix A).

### **3.3.3**

Acute phase serum samples were isolated from Chikungunya virus (see Appendix B, B.1).

### **3.3.4**

Chikungunya virus nucleic acid was detected in acute phase serum samples (see B.2 in Appendix B).

## **4 Diagnostic principles**

Comprehensive judgment based on the patient's epidemiological history, clinical manifestations and laboratory findings. Has never happened in Chikungunya fever the area should be based on laboratory inspections.

### **5.1 Suspected cases**

Meets 3.1, 3.2.1, 3.2.2, and/or 3.2.3 and excludes dengue and/or other fever with rash.

### **5.2 Clinical diagnosis cases**

Meets 5.1 and complies with 3.3.1.

### **5.3 Confirmed cases**

Meets 5.1 or 5.2 and complies with any of 3.3.2, 3.3.3, 3.3.4.

## **6 Differential diagnosis**

Chikungunya fever should be associated with dengue fever, alphavirus infection, infectious erythema, post-infection arthritis (including rheumatic fever), Scarlet fever, rickettsial disease (typhus, ascariasis), measles, drug eruption, etc. See Appendix C and Appendix D.

Appendix A

(normative appendix) Chikungunya fever serological test method A.1 Enzyme-linked immunosorbent assay for chikungunya virus (CHIKV) IgM antibody (IgM antibody capture enzyme linked Immunosorbent assay A.1.1 Principle First, a 96-well plate is coated with an anti-human IgM  $\mu$ -chain antibody (capture antibody), and if a CHIKV anti-IgM antibody is present in the serum to be tested, the captured antibody binds and then binds to the CHIKV antigen, and finally binds to the enzyme-labeled anti-CHIKV antibody, and the enzyme interacts with the chromogenic substrate.

A color reaction will occur with the use.

A.1.2 Materials and reagents Materials and reagents are as follows.

a) coating solution. sodium carbonate/sodium bicarbonate buffer, pH 9.6, 1.59 g Na<sub>2</sub>CO<sub>3</sub>

and 2.93 g NaHCO<sub>3</sub> dissolved in 1 L of water;

- b) lotion. phosphate buffered saline (PBS), containing 0.05% Tween 20, pH 7.2;
- c) blocking solution. PBS containing 5% milk powder and 0.05% Tween 20;
- d) coated antibody. anti-human IgM $\mu$  chain antibody;
- e) viral antigen. inactivated purified CHIKV antigen, non-infectious;
- f) cell antigen. purified uninfected virus cell culture medium;
- g) enzyme-labeled antibody. horseradish peroxidase-labeled CHIKV monoclonal antibody;
- h) enzyme substrate. TMB;
- i) Stop solution. 2N H<sub>2</sub>SO<sub>4</sub>;
- j) Clinical specimens. serum and/or cerebrospinal fluid specimens from patients with acute phase; positive and negative serum controls;
- k) Equipment. flat bottom 96-well plate, plate washer, microplate reader, incubator, single-channel and multi-channel pipettes, reagent reservoir, fresh-keeping Bag, absorbent paper.

A.1.3 Detection method Follow these steps.

- a) Mark the experiment number on the plate with a marker to determine the location of each clinical specimen.
  - b) Dilute the anti-human IgM $\mu$  chain antibody in a 1:2000 ratio with a pH 9.6 coating and add to the well of a 96-well plate at 75  $\mu$ L/well.
- Incubate overnight at 4 °C.
- c) Pour off the antibody coating and blot the remaining liquid on the plate to blotting paper. Add 200  $\mu$ L of blocking solution to each well to block the reaction plate. room Incubate for 30 min at room temperature.
  - d) Wash the plate 5 times with the washing machine on the automatic washer. Make sure that each well is filled with lotion for each wash.
  - e) The patient's serum is diluted 1:100 with a dilution, and the cerebrospinal fluid is diluted 1:10, then 50  $\mu$ L/well, 2 wells each.

At the same time, the positive control serum and the negative control serum were diluted 1:100 with a diluent, and 2 wells were added for each. Incubate for 1 h in a 37 °C wet box.

Wash 5 times.

- f) Dilute viral antigens and cellular antigens. Add virus antigen to 50  $\mu$ L/well on the left side of each row, and 50  $\mu$ L/well on the right side of each row.