



**Maldives**  
**Health Protection Agency**  
**Ministry of Health**

# **Guidelines for sending samples to test for Chikungunya/Zika**

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**Health Protection Agency**  
**Ministry of Health**

Maldives being an endemic country for the Aedes mosquito, we are at risk of experiencing outbreaks of Dengue, Chikungunya and Zika. This is a guideline for healthcare professionals on how and when to send samples for further testing if Chikungunya or Zika is suspected. We will be using the Trio plex Real-time RT-PCR Assay to test for Dengue/Chikungunya/Zika to test for Chikungunya and Zika. The testing is part of surveillance only and is not to be used as a diagnostic test. Treatment for all the 3 diseases should be based on the clinical diagnosis and its treatments guidelines.

### 1. Sample collection criteria:

For the purposes of this surveillance, the case definition to be used to recruit patients for sample collection is:

- Those who are negative for the NS1 antigen (Ag) rapid test for Dengue virus.
- Patient who cannot be clinically diagnosed as Chikungunya but those who cannot be eliminated either.

Given below are the specific signs and symptoms of Chikungunya.

- fever ( $> 37.8^{\circ}\text{C}$ ) OR maculopapular rash  
with one or more of the following (not explained by any other conditions)
- arthralgia
- myalgia
- non-purulent conjunctivitis
- conjunctival hyperemia
- headache
- fatigue/malaise

Note: Those who are clinically diagnosed as Chikungunya **MUST** be reported just like any other notifiable disease.

The samples taken will be used for surveillance purposes only.

### 2. Samples:

Blood should be collected during acute phase (within the first 6 days of symptom onset). Samples collected outside IGMH should be sent to IGMH laboratory within 24 hours. Further guidance is given in Appendix 1- Guidance for Sample Collection, Processing and Shipment.

### 3. Labeling:

Labeling of the sample should be as per the usual sample labeling guidelines followed by the laboratory.

### 4. Procedure:

An information sheet containing the following information should be sent with the sample.

- The patient name, age, gender, ID card number/passport number, contact number and residential address, permanent address, clinical history, date of onset of symptoms .
- Name of the person who collected the sample, designation, date of sample collection and the time of sample collection.

## 5. Guidance for the specimen collection processing and shipment

### 1. Specimen collection and processing

#### 1.1. Serum specimen

- 1.1.1. The serum sample should be obtained during the acute phase of the infection (the first 6 days of illness).
- 1.1.2. All acute specimens will be tested by RT-PCR; virus isolation will be attempted on positive samples.

Type of sample	Interval since the onset of symptoms	Type of analysis
Acute	Until day 6	RT-PCR

- 1.1.3. Collect blood sample ( $\approx 3$  ml) in a blood collection tube.
- 1.1.4. After the sample collection, the blood is allowed to clot for 30-60 minutes at room temperature. Blood tubes should then be stored at 2-8 °C, (in refrigerator, wet ice, or with ice pack) until arrival at IGMH laboratory for centrifugation.

**Note: Sample can be stored at 2-8 °C, in refrigerator, on wet ice or ice pack for no longer than 72 hours before centrifugation.**

AT IGMH

- 1.1.5. Separate serum by centrifugation at 1000-1500 G-Force or Relative Centrifugation Force (RCF) or 1,800 Revolutions per minute (rpm) at 2-8 °C or room temperature.

**Note:**

- For the centrifuge with swinging bucket, centrifuge for 10 min.
- For the centrifuge with fixed-angle, centrifuge for 15 min.

**Note:**

- 1. Sample must be stored at 2-8 °C, in refrigerator, on wet ice or ice pack for no longer than 24 hours.**
- 2. For longer than 24 hours, sample must be stored at - 20  $\pm$  10 °C or dry ice until arrival at IGMH laboratory.**