

# TaqMan™ Arbovirus Triplex Kit (ZIKV/DENV/CHIKV), 0.2-mL Block

Lyophilized reagents for multiplex real-time RT-PCR detection of Zika, Dengue, and Chikungunya virus RNA

Catalog Number A31746

Pub. No. MAN0016006 Rev. D00



**WARNING!** Read the Safety Data Sheets (SDSs) and follow the handling instructions. Wear appropriate protective eyewear, clothing, and gloves. Safety Data Sheets (SDSs) are available from [thermofisher.com/support](https://www.thermofisher.com/support).

## Product description

The TaqMan™ Arbovirus Triplex Kit (ZIKV/DENV/CHIKV) is designed to detect viral RNA, prepared from urine or serum research samples, for the following viruses:

- Zika virus with Asian lineage
- Dengue virus from 4 serotypes: DENV-1, DENV-2, DENV-3, and DENV-4
- Chikungunya virus

The kit also detects human endogenous control PPIA (Cyclophilin A), to monitor nucleic acid recovery and to serve as a process control for the RT-PCR.

The kit includes primers and TaqMan™ probes for the viral and PPIA targets, and other reagents for RT-PCR, in a lyophilized format. After addition of RNA sample, the reconstituted reagents are ready for real-time RT-PCR.

## Contents and storage

**Table 1** TaqMan™ Arbovirus Triplex Kit (ZIKV/DENV/CHIKV) (Cat. No. [A31746](#); 96 reactions)

Contents	Amount	Storage
Lyophilized assay, Standard, 0.2-mL tube	12 × 8-tube strips	<ul style="list-style-type: none"> <li>• 18–28°C for up to 3 years<sup>[1]</sup></li> <li>• 2–8°C for long-term storage</li> <li>• Protect from moisture<sup>[2]</sup></li> </ul>
MicroAmp™ Optical 8-Cap Strips	12 × 8-cap strips	Room temperature

<sup>[1]</sup> Product is shipped at ambient temperature. See [thermofisher.com/ambientshipping](https://www.thermofisher.com/ambientshipping).

<sup>[2]</sup> See "Procedural guidelines" on page 2.

## Required materials

Unless otherwise indicated, all materials are available through [thermofisher.com](https://www.thermofisher.com). "MLS" indicates that the material is available from [fisherscientific.com](https://www.fisherscientific.com) or another major laboratory supplier.

Item	Source
<b>Applied Biosystems™ real-time PCR instrument and accessories, one of the following:</b>	
QuantStudio™ instrument capable of detecting at least 5 colors <sup>[1]</sup> : <ul style="list-style-type: none"> <li>• QuantStudio™ 5 Real-Time PCR System</li> <li>• QuantStudio™ 12K Flex Real-Time PCR System</li> <li>• QuantStudio™ 6 / QuantStudio™ 7 Flex Real-Time PCR System</li> </ul>	Contact your local sales office
7500 Real-Time PCR Instrument Precision Plate Holder for 0.2 mL Tubes and Strips (4367033)	Contact your local sales office
<b>Equipment</b>	
MicroAmp™ 96-Well Base	<a href="#">N8010531</a>
MicroAmp™ Cap Installing Tool	<a href="#">4330015</a>
Benchtop microcentrifuge with 8-tube strip adapter, or plate centrifuge	MLS
Laboratory mixer, Vortex or equivalent	MLS
Adjustable pipettors	MLS
<b>Plastics and consumables</b>	
(Optional) MicroAmp™ Optical 8-Cap Strips <sup>[2]</sup>	<a href="#">4323032</a>
(Optional) MicroAmp™ 8-Tube Strip, 0.2 mL <sup>[2]</sup>	<a href="#">N8010580</a>
Aerosol-resistant micropipette tips	MLS
Disposable gloves	MLS
<b>Reagents</b>	
Nuclease-free water	<a href="#">AM9938</a>

<sup>[1]</sup> Precision Plate Holder is included with the instrument.

<sup>[2]</sup> Required only for the 7500 series instrument, to balance the lid pressure if less than 2 full strips are processed.

## Procedural guidelines

- Protect the lyophilized assay from moisture; ambient moisture will compromise performance very quickly. Use multiple barriers.  
For example, after the original pouch is opened:
  - Place unused strips in the original pouch with the silica desiccant pack, then seal the pouch. Use a resealable bag if the original pouch is broken.
  - Place the sealed pouch in a dry box or desiccator.
- Do not use DEPC-treated water.
- Ensure that personnel operating the real-time PCR instrument are trained.
- Ensure that the instrument is calibrated for each detector dye and passive reference dye, according to the instrument user guide. See “Dye spectral calibration plates” on page 2.
- Ensure that the appropriate Precision Plate Holder is installed in the instrument. Follow the instrument user guide for tube placement and plate holder use.

## Guidelines for input RNA

Use high-quality RNA samples for reliable PCR results.

**Table 2 Recommended RNA isolation kits**

Kit	Cat. No.	Notes
MagMAX™ Pathogen RNA/DNA Kit	4462359	Use up to 25 µL of total RNA in elution buffer per PCR reaction.
PureLink™ Viral RNA/DNA Mini Kit	12280050	

## Set up and run the reactions

- If necessary, adjust the total volume of RNA sample to 25 µL per reaction, using nuclease-free water.
- Remove the cap of the 8-tube strip; discard the cap.
- Add 25 µL of RNA sample to each tube, then firmly apply a new optical cap strip (provided in the kit).
- Mix by flicking the tube strip several times or by vortexing briefly, then centrifuge briefly.
- Select or create dye detectors, then assign to each tube in the layout.

Target	Reporter	Quencher
Zika	FAM™ dye	Non-fluorescent quencher (NFQ)
Dengue	VIC™ dye	
Chikungunya	ABY™ dye	
PPIA	JUN™ dye	

- Load the tube strips and run the real-time PCR instrument using the following thermal cycling conditions.
  - Run mode: Fast

- Passive reference: MUSTANG PURPLE™ dye.

Stage	Cycles	Temperature	Time
Reverse transcription	1	50°C	20 minutes
Activation	1	95°C	2 minutes
Amplification	40	95°C	15 seconds
		60°C	1 minute

## Guidelines for data analysis

The general process for data analysis is to:

- View the amplification plots.
- Set the baseline and threshold values.
- Use the instrument software to calculate C<sub>t</sub> values.

Expected results:

- Amplification should not be seen in no-template control (NTC) reactions.
- Amplification of the PPIA target should be seen in samples with human RNA present.
- Amplification of the Zika, Dengue, or Chikungunya target should be seen in samples when viral RNA is present.

## Dye spectral calibration plates

See your instrument user guide for recommended calibration schedules and detailed calibration instructions.

Calibration plates	Cat. No.
QuantStudio™ 3/5 10-Dye Spectral Calibration Kit	A26343
QuantStudio™ 3/5 Spectral Calibration Plate 1	A26331
QuantStudio™ 3/5 Spectral Calibration Plate 2	A26332
7500 Real-Time PCR Systems Spectral Calibration Kit I	4349180
ABY™ Dye Spectral Calibration Plate	A24738
FAM™ Dye Spectral Calibration Plate	4432327
JUN™ Dye Spectral Calibration Plate	A24737
MUSTANG PURPLE™ Dye Spectral Calibration Plate	4461599
VIC™ Dye Spectral Calibration Plate	4432334

## Limited product warranty

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Revision history: Pub. No. MAN0016006 D00

Revision	Date	Description
D00	25 March 2024	<ul style="list-style-type: none"><li>• The legal manufacturer was updated.</li><li>• The shelf life was updated to 3 years.</li><li>• Dye and calibration plate products were updated.</li></ul>
C.0	17 August 2022	Changed the kit name from Zika Virus to Arbovirus throughout.
B.0	16 April 2018	<ul style="list-style-type: none"><li>• Updated the targets that have expected amplification.</li><li>• Update license information.</li></ul>
A.0	29 August 2016	New document.

The information in this guide is subject to change without notice.

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