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SUPPORTING INFECTIOUS DISEASE RESEARCH

# **Dengue Virus Type 4 Primers**

## Catalog No. NR-12081

This reagent is the tangible property of the U.S. Government.

## For research use only. Not for use in humans.

#### **Contributor and Manufacturer:**

**BEI Resources** 

#### **Product Description:**

There are four antigenically related but genetically distinct serotypes of dengue virus, types 1 through 4 (DENV-1 to DENV-4, respectively). NR-12081 contains forward and reverse primers that specifically amplify a region of the NS2A gene of DENV-4. A protocol is outlined in Appendix I.

#### **Material Provided:**

Each vial contains approximately 100  $\mu$ L of a mixture of forward and reverse primers in TE buffer (pH 7.0). The concentration is shown on the Certificate of Analysis.

### Packaging/Storage:

Primers were packaged aseptically in screw-capped plastic cryovials. The product is provided frozen on dry ice and should be stored at -60°C or colder upon arrival. Freeze-thaw cycles should be minimized.

#### Citation:

Acknowledgment for publications should read "The following reagent was obtained through BEI Resources, NIAID, NIH: Dengue Virus Type 4 Primers, NR-12081."

#### **Biosafety Level: 1**

Appropriate safety procedures should always be used with this material. Laboratory safety is discussed in the following publication: U.S. Department of Health and Human Services, Public Health Service, Centers for Disease Control and Prevention, and National Institutes of Health. <u>Biosafety in Microbiological and Biomedical Laboratories (BMBL)</u>. 6th ed. Washington, DC: U.S. Government Printing Office, 2020.

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# APPENDIX I

# Dengue Virus Type 4 (DENV-4) Primers

# **Recommended Reagents/Equipment**

Reagent	Source	Catalog #
DENV-4 Primers	BEI Resources	NR-12081
Positive Control Template, Genomic RNA from DENV-4, H241 (tissue culture adapted)	BEI Resources	Genomic RNA from NR-86
Qiagen OneStep RT-PCR Kit	Qiagen	210212

### **Reaction Mix<sup>1</sup>**

Reagent	Stock Concentration	Volume per Reaction (µL)
RNase-free water		23
Qiagen OneStep RT-PCR Buffer	5X	10
Q Solution	5X	10
dNTP Mix	10 mM each	2
Qiagen OneStep RT-PCR Enzyme Mix		2
Primers <sup>2</sup>	10 µM (each primer)	1
Template	50 to 5000 ng	2
		Total – 50 μL

<sup>1</sup>Reaction mix should be kept on bench-top cooler until ready for use. <sup>2</sup>Primers are supplied at working stock concentrations.

#### Cycling Protocol

Cycle	# of Repeats	Step	Conditions	
1	1	1	54°C for 30 minutes	
1	1	1	95°C for 15 minutes	
2 35		1	94°C for 30 seconds	
	35	2	54°C for 30 seconds	
		3	72°C for 2 minutes	
3	1	1	72°C for 7 minutes	
4	Indefinite	1	Hold at 4°C	