

pCR®II-TOPO® Containing Hemagglutinin (HA) Gene from Monkeypox Virus, Zaire 79, Linearized

Catalog No. NR-4076

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Contributor:

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Product Description:

The hemagglutinin (HA) gene from monkeypox virus, Zaire 79 (BEI Resources NR-2324) was amplified from extracted DNA and cloned into the phagemid vector, pCR®II-TOPO®. The plasmid was produced in *Escherichia coli* DH5 α ™-T1^R cells, extracted using a QIAGEN Plasmid Maxi Kit and linearized with *Hind*III. The sequence of the HA gene of NR-4076 is shown in Table 1 and differs from the monkeypox virus, Zaire 79 HA sequence (GenBank: DQ011155)¹ by one nucleotide at position 516 (T to C).

NR-4076 has been qualified for use as a standard for real-time quantitative PCR. A Monkeypox Virus Hemagglutinin Gene-Specific Quantitative Real-Time PCR Assay Detection Kit containing forward and reverse primers (BEI Resources NR-9348 and NR-9349), probe (BEI Resources NR-9347) and NR-4076 is available as BEI Resources NR-9351.

Material Provided:

Each vial contains approximately 0.14 μ g of plasmid DNA in TE buffer (10 mM Tris-HCl, 1 mM EDTA, pH 7.0). The concentration is shown on the Certificate of Analysis.

Packaging/Storage:

NR-4076 was packaged aseptically in screw-capped plastic cryovials. The product is provided frozen on dry ice and should be stored at -60°C or colder immediately upon arrival. Freeze-thaw cycles should be minimized.

Citation:

Acknowledgment for publications should read "The following reagent was obtained through the NIH Biodefense and Emerging Infections Research Resources Repository, NIAID, NIH: pCR®II-TOPO® Containing the Hemagglutinin (HA) Gene from Monkeypox Virus, Zaire 79, Linearized, NR-4076."

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. Biosafety in Microbiological and Biomedical Laboratories, 5th ed. Washington, DC: U.S. Government Printing Office, 2007; see www.cdc.gov/od/ohs/biosfty/bmb15/bmb15toc.htm.

Disclaimers:

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References:

1. Likos, A. M., et al. "A Tale of Two Clades: Monkeypox Viruses." *J. Gen. Virol.* 86 (2005): 2661–2672. PubMed: 16186219. GenBank: DQ011155.
2. Hooper, J. W., D. M. Custer, and E. Thompson. "Four-Genes-Combination DNA Vaccine Protects Mice against a Lethal Vaccinia Virus Challenge and Elicits Appropriate Antibody Responses in Nonhuman Primates." *Virology* 306 (2003): 181–195. PubMed: 12620810.

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Table 1. HA Gene and Flanking Sequences in NR-4076

ATAGGGCGAATTGGGCCCTCTAGATGCATGCTCGAGCGGCCGCCAGTGTG
ATGGATATCTGCAGAATTCGCCCTT**ATTTGGAAAGTTTTATAGGTAGTTG**
ATAGAACAAAATACATAATTTTGTAAAAATAAATCACTTTTTATACTAAT
ATGACACAATTACCAATACTTTTGTACTAATATCATTAGTATACGCTAC 50
 ACCTTCTCCTCAGACATCTAAAAAATAGGTGATGATGCAACTATATCAT 100
 GTAGTCGAAATAATACAAATTACTACGTTGTTATGAGTGCTTGGTATAAG 150
 GAGCCCAATTCCATTATTCTCTTAGCTGCCAAAAGCGACGTCTTGTATTT 200
 TGATAATTATACCAAGGATAAAATATCTTACGACTCTCCATACGATGATC 250
 TAGTTACAACATCACAATTAATCATTGACTGCTGGAGATGCCGGTACT 300
 TATATATGTGCATTCTTTATGACATCGACTACAAATGATACTGATAAAGT 350
 AGATTATGAAGAATACTCCATAGAGTTGATTGTAATAACAGATAGTGAAT 400
 CGACTATAGACATAATACTATCTGGATCTACACCGAAACTATTTCTGAG 450
 AAACCAGAGGATATAGATAATTCTAATTGCTCGTCTGTATTTCGAAATCGC 500
 GACTCCGGAACCAATCACTGATAATGTAGAAGACCATACAGACACCGTCA 550
 CATACTAGTGATAGCATTAATACAGTAAATGCATCATCTGGAGAATCC 600
 ACAACAGACGAGACTCCGGAACCAATTACTGATAAAGAAGAAGATCATAAC 650
 AGTAACAGACACTGTCTCATACTACAGTAAGTACATCATCTGGAATTG 700
 TCACTACTAAATCAACCACCGATGATGCGGATCTTTATGATACATACAAT 750
 GATAATGATACAGTACCGCAACTACTGTAGGTGGTAGTACAACCTCTAT 800
 TAGCAATTATAAAACCAAGGACTTTGTAGAAATATTTGGTATTACCACAT 850
 TAATTATATTGTCGGCAGTGGCGATCTTCTGTATTACGTATTATATATGT 900
 AATAAACACCCACGTAAATACAAAACAGAGAACAAAGTCT**TAGATTTTTGA**
 CTTACATAAAATATCTGGGATAATAAAATCTATCATATTGAGAGGACCATC
 TGGTTTCAGGAA**AGACAGCCATAAACCAAAAAGACT**AAGGGCGAATTCCAGCA
 CACTGGCGGCCGTTACTAGTGGATCCGAGCTCGGTACCA**AAGCTT**GATGCA
TAGCTTGAGTATTCTATAGTGTACCTAAA

The monkeypox virus HA gene sequence is numbered and the ATG and TAG codons are bolded. The nucleotide difference from GenBank DQ011155 at position 516 of the HA gene is highlighted in grey. Flanking monkeypox virus gene sequences are italicized. Plasmid sequences are underlined. The *Hind*III site is underlined and bolded. Primer sequences used to amplify the HA gene for cloning are highlighted in yellow.