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Technical Data Sheet:

Quantitative Synthetic Influenza A Virus (H1N1) RNA

ATCC® Number	VR-3386SD™
Product Description	Quantitative Synthetic Influenza A virus (H1N1) RNA is a synthetically derived preparation that can be used for assay development, verification, and validation as well as monitoring of day-to-day test variation and lot-to-lot performance of molecular-based assays. The quantitative format allows for the generation of a standard curve for quantitative PCR (qPCR) to determine viral load.
Genetic Target	The synthetic RNA preparation includes two constructs. One construct includes the full genes for the HA and NP regions. The other construct includes the full genes for the NA, M1/M2, and NEP/NS1 regions. This product is based on the A/Brisbane/59/2007 (H1N1) influenza virus sequence with few modifications to accommodate manufacturing and product compatibility with diagnostically relevant assays.

Publication	Assay Target	Oligo	Sequence (5' to 3')	Number of mismatches with ATCC® VR-3386SD™ based on *in silico* analysis
World Health Organization. WHO information for the molecular detection of influenza viruses. Publish date: February 2021.	HA	Forward	CCCAGGGYATTTCGCYGACTATGAG	0
		Reverse	CATGATGCTGAYACTCCGGTTACG	0
		Probe	TCTCAAAYGAAGATACTGAACT	0
Hoffmann B, et al. Riems influenza a typing array (RITA): An RT-qPCR-based low density array for subtyping avian and mammalian influenza a viruses. Sci Rep 6: 27211, 2016. PubMed: 27256976	HA	Forward	ACACAATATGTATAGGYTAHCATGC	0
		Reverse	GAGTGTGTYACTGTYACATTCTT	0
		Probe	TCDACMGACACTGTWGACACAGTACTNGA	0

Hoffmann B, et al. Riems influenza a typing array (RITA): An RT-qPCR-based low density array for subtyping avian and mammalian influenza a viruses. Sci Rep 6: 27211, 2016. PubMed: 27256976	HA	Forward	CCCGTCYATTCAATCYAGAGG	0
		Reverse 1	GGTGATAACCRTACCANCCATC	1
		Reverse 2	TCATTTTGATGRTGATAACCRTACCA	1
		Probe	CATYCCWGTCCAYCCYCCTTCAATGAA	0
Goecke NB, et al. Subtyping of Swine Influenza Viruses Using a High-Throughput Real-Time PCR Platform. Front Cell Infect Microbiol 8: 165, 2018. PubMed: 29872645	HA	Forward	GGWTGGTATGGTTATCATCAT	0
		Reverse	CTCGATTACAGAGTTCACC	0
		Probe	CAGGGATCTGGCTATGCTGCAGAYC	1
Haach V, et al. One-step multiplex RT-qPCR for the detection and subtyping of influenza A virus in swine in Brazil. J Virol Methods 269: 43-48, 2019. PubMed: 30959063	HA	Forward	GGTTTGTTTGGWGCCATTGC	0
		Reverse	CAGCATAVCCAGAYCCTTGC	0
		Probe	TTCATTGAAGGRGGDTGGACTGGAAT	0
Suwannakarn K, et al. Typing (A/B) and subtyping (H1/H3/H5) of influenza A viruses by multiplex real-time RT-PCR assays. J Virol Methods 152(1-2): 25-31, 2008. PubMed: 18598722	HA	Forward	ACTACTGGACTCTGCTKGAA	0
		Reverse	AAGCCTCTACTCAGTGCGAA	0
		Probe	TTGAGGCAAATGGAAATCTAATAGC	0
Centers for Disease Control and Prevention (U.S.); National Center for Immunization and Respiratory Diseases (U.S.). Influenza Division. Virology Surveillance and Diagnosis Branch. Genomics and Diagnostics Team. Research Use Only CDC Influenza SARS-CoV-2 (Flu SC2) Multiplex Assay Real-Time RT-PCR Primers and Probes. Publish date: July 14, 2020.	M	Forward 1	CAAGACCAATCYTGTCACCTCTGAC	0
		Forward 2	CAAGACCAATYCTGTCACCTYTGAC	0
		Reverse 1	GCATTYTGGACAAAVCGTCTACG	0
		Reverse 2	GCATTTTGGATAAAGCGTCTACG	1
		Probe	TGCAGTCCTCGCTCACTGGGCACG	0
World Health Organization. WHO information for the molecular detection of influenza viruses. Publish date: February 2021.	M	Forward	GACCRATCCTGTCACCTCTGAC	0
		Reverse	AGGGCATTYTGGACAAAKCGTCTA	0
		Probe	TGCAGTCCTCGCTCACTGGGCACG	0
World Health Organization. WHO information for the molecular detection of influenza viruses. Publish date: February 2021.	M	Forward	ATGAGYCTTYTAACCGAGGTCGAAACG	0
		Reverse	TGGACAAANCGTCTACGCTGCAG	0
World Health Organization. WHO information for the molecular detection of influenza viruses. Publish date: February 2021.	M	Forward	CTTCTAACCGAGGTCGAAACGTA	0
		Reverse	GGTGACAGGATTGGTCTTGTCTTTA	0
		Probe	TCAGGCCCCCTCAAAGCCGAG	0

World Health Organization. WHO information for the molecular detection of influenza viruses. Publish date: February 2021.	M	Forward	CCMAGGTCGAAACGTAYGTTCTCTCTATC	1
		Reverse	TGACAGRATYGGTCTTGTCTTTAGCCAYTCCA	0
		Probe	ATYTCGGCTTTGAGGGGGCCTG	0
Spackman E, et al.Development of a Real-Time Reverse Transcriptase PCR Assay for Type A Influenza Virus and the Avian H5 and H7 Hemagglutinin Subtypes. J Clin Microbiol 40(9): 3256-3260, 2022. PubMed: 12202562	M	Forward	AGATGAGTCTTCTAACCGAGGTCG	0
		Reverse	TGCAAAAACATCTTCAAGTCTCTG	0
		Probe	TCAGGCCCCCTCAAAGCCGA	0
Laconi A, et al. Detection of avian influenza virus: a comparative study of the in silico and in vitro performances of current RT-qPCR assays. Sci Rep 10(1): 8441, 2020. PubMed: 32439885	M	Forward	AGATGAGTCTTCTAACCGAGGTCG	0
		Reverse	TGCAAARACATCTTCAAGTYTCTG	0
		Probe	TCAGGCCCCCTCAAAGCCGA	0
Laconi A, et al. Detection of avian influenza virus: a comparative study of the in silico and in vitro performances of current RT-qPCR assays. Sci Rep 10(1): 8441, 2020. PubMed: 32439885	M	Forward	AGATGAGYCTTCTAACCGAGGTCG	0
		Reverse	TGCAAANACATCYTCAAGTCTCTG	0
		Probe	TCAGGCCCCCTCAAAGCCGA	0
Liu J, et al. Development and application of a triplex real-time PCR assay for the simultaneous detection of avian influenza virus subtype H5, H7 and H9. J Virol Methods 252: 49-56, 2018. PubMed: 29129489	M	Forward	GACCAATCCTGTCACCTCTGAC	0
		Reverse	GGGCATTTTGGACAAAGCGTCTACG	0
Nagy A, et al. A universal RT-qPCR assay for “One Health” detection of influenza A viruses. PLoS One 16(1): e0244669, 2021. PubMed: 33471840	M	Forward	GGCCCCCTCAAAGCCGA	0
		Reverse	CGTCTACGYTGCAGTCC	0
		Probe	TCACTKGGCACGGTGAGCGT	0
Goecke NB, et al. Subtyping of Swine Influenza Viruses Using a High-Throughput Real-Time PCR Platform. Front Cell Infect Microbiol 8: 165, 2018. PubMed: 29872645	M	Forward	CTTCTAACCGAGGTCGAAACGTA	0
		Reverse	CACTGGGCACGGTGAGC	0
		Probe	TCAGGCCCCCTCAAAGCCGA	0
Hassan KE, et al. Improved Subtyping of Avian Influenza Viruses Using an RT-qPCR-Based Low Density Array: ‘Riems Influenza a Typing Array’, Version 2 (RITA-2). Viruses 14(2): 415, 2022. PubMed: 35216008	M	Forward	AGATGAGYCTTCTAACCGAGGTCG	0
		Reverse 1	TGCAAAAACATCTTCAAGTYTCTG	0
		Reverse 2	TGCAAAIACATCYTCAAGTYTCTG	1
		Probe	TCAGGCCCCCTCAAAGCCGA	0

Leong NKC, et al. A six-plex droplet digital RT-PCR assay for seasonal influenza virus typing, subtyping, and lineage determination. Influenza Other Respir Viruses 14(6): 720-729, 2020. PubMed: 32519796	M	Forward	CTTCTAACCGAGGTCGAAACGTA	0
		Reverse	AGGGCATTYTGGACAAAKCGTCTA	0
		Probe	TCAGGCCCCCTCAAAGCCGAG	0
Suwannakarn K, et al. Typing (A/B) and subtyping (H1/H3/H5) of influenza A viruses by multiplex real-time RT-PCR assays. J Virol Methods 152(1-2): 25-31, 2008. PubMed: 18598722	M	Forward	CATGGARTGGCTAAAGACAAGACC	0
		Reverse	AGGGCATTTTGGACAAAKCGTCTA	0
		Probe	ACGC+TCACCG+TGCCC+AGT	0
Van Elden LJ, et al. Simultaneous detection of influenza viruses A and B using real-time quantitative PCR. J Clin Microbiol 39(1): 196-200, 2001. PubMed: 11136770	M	Forward	GGACTGCAGCGTAGACGCTT	0
		Reverse 1	CATCCTGTTGTATATGAGGCCCAT	0
		Reverse 2	CATTCTGTTGTATATGAGGCCCAT	1
		Probe	CTCAGTTATTCTGCTGGTGCACTTGCCA	0
Ward CL, et al. Design and performance testing of quantitative real time PCR assays for influenza A and B viral load measurement. J Clin Virol 29(3): 179-188, 2004. PubMed: 14962787	M	Forward	AAGACCAATCCTGTCACCTCTGA	0
		Reverse	CAAAGCGTCTACGCTGCAGTCC	0
		Probe	TTTGTGTTCACGCTCACCGT	0
World Health Organization. WHO information for the molecular detection of influenza viruses. Publish date: February 2021.	NA	Forward	AGACCTTGCTTCTGGGTTGA	0
		Reverse	ACCGTCTGGCCAAGACCA	0
		Probe	ATCTGGACTAGCGGGAGCAGCAT	1
World Health Organization. WHO information for the molecular detection of influenza viruses. Publish date: February 2021.	NA	Forward	TGGATGGACAGATACCGACA	0
		Reverse	CTCAACCCAGAAGCAAGGTC	0
		Probe	CAGCGGAAGTTTCGTTCAACAT	0
Hoffmann B, et al. Riems influenza a typing array (RITA): An RT-qPCR-based low density array for subtyping avian and mammalian influenza a viruses. Sci Rep 6: 27211, 2016. PubMed: 27256976	NA	Forward	AGRCCTTGYTTCTGGGTTGA	0
		Reverse	ACCGTCTGGCCAAGACCA	0
		Probe	ATYTGGACYAGTGGGAGCAGCAT	0
Goecke NB, et al. Subtyping of Swine Influenza Viruses Using a High-Throughput Real-Time PCR Platform. Front Cell Infect Microbiol 8: 165, 2018. PubMed: 29872645	NA	Forward	AGRCCTTGYTTCTGGGTTGA	0
		Reverse	ACCGTCTGGCCAAGACCA	0
		Probe	ATYTGGACYAGTGGGAGCAGCAT	0
Hassan KE, et al. Improved Subtyping of Avian Influenza Viruses Using an RT-qPCR-Based Low Density Array: 'Riems Influenza a Typing Array', Version 2 (RITA-2). Viruses 14(2): 415, 2022. PubMed: 35216008	NA	Forward	GRCCTTGYTTCTGGGTKGA	0
		Reverse	ACCGTCTGGCCAAGACCA	0
		Probe	CAATYTGGACYAGTGGRAGYAGCAT	0