

TaqMan® AIV-Matrix and Xeno™ RNA Controls

Do not store in a frost-free freezer.

Catalog # (P/N): 4405544

Materials Provided:	Component	Amount	Storage
	Xeno™ RNA Control (10,000 copies/μL)	110 μL	-20°C
	25X AIV-Matrix Control RNA (10,000 copies/μL)	15 μL	-20°C
	Nucleic Acid Dilution Solution	500 μL	-20°C

Safety Information: Read the MSDS, and follow the handling instructions. Wear appropriate protective eyewear, clothing, and gloves.

USER INFORMATION

Applications:

Using Xeno RNA Control

Xeno RNA Control is an internal, positive control target. Add Xeno RNA Control to the sample lysis solution used for RNA isolation, to serve as a positive control both for recovery of RNA and for the RT-PCR.

Add 5,000–10,000 copies Xeno RNA Control per isolation (0.5–1 μL at 10,000 copies/μL).

Using AIV-Matrix Positive Control

Include duplicate positive control reactions with the AIV-Matrix Control RNA. Use 1 μL of 25X AIV-Matrix Control RNA per reaction.

Negative controls for RT-PCR

Include duplicate no-sample control reactions that contain Nuclease-free Water instead of sample or positive control RNA.

Control and experimental RT-PCRs (25 μL reactions)

Reaction Type	Sample	Amount
Experimental RNA	RNA	8.83 μL
No-sample control	Nuclease-free Water	8.83 μL
Positive control	25X AIV-Matrix Control RNA	1 μL
	Nuclease-free Water	7.83 μL

Troubleshooting:

See below for general troubleshooting tips.

No signal from Xeno RNA Control

Lack of signal may indicate that the RNA contains inhibitors of RT and/or PCR. Increasing the number or stringency of washes during RNA isolation may help to eliminate inhibitors from subsequent samples.

Samples containing minimal amounts of inhibitors may yield successful RT-PCRs by adding less sample (and therefore less inhibitor), to the reaction. Alternatively, samples can be diluted, for example 5- and 10-fold, and then used in RT-PCR.

Evaluating problems with RNA isolation

If Xeno RNA Control was added to the lysis solution used for RNA isolation, a lack of signal from both the target and Xeno RNA Control indicates either a problem with RNA recovery or that samples contain inhibitors of RT-PCR.

- If carrier RNA was used in viral RNA isolation from cell-free samples, check its concentration to evaluate its recovery.
- If Xeno RNA Control was added to the lysis solution used for RNA isolation, compare the result of amplifying sample RNA using RT-PCR master mixes with and without Xeno RNA Control at 200 copies/reaction.
 - If the reaction amplified using master mix with Xeno RNA Control results in signal, but the reaction amplified using master mix without Xeno RNA Control does not result in signal, this indicates that the Xeno RNA Control was not recovered.
 - If Xeno RNA Control signal is not seen from either sample, this indicates that the RNA contains inhibitors of RT-PCR (see above for troubleshooting suggestions).

No signal from AIV-Matrix Control RNA (positive control reaction)

If the positive control reaction containing the AIV-Matrix Control RNA fails to produce an RT-PCR signal, the reaction may have failed. Potential causes for reaction failure include the following:

- Improper handling of the AIV-Matrix Control RNA, resulting in RNA degradation. As with any RNA sample, it is important to use typical precautions against RNase contamination when handling the Control RNA. For example, we recommend wearing clean gloves and using nuclease-free barrier pipet tips.
- The thermal cycler was not properly programmed.
- The RT-PCR master mix reagents were handled improperly, lost activity, or were set up incorrectly.

Signal detected in the no-sample control

The most likely cause of signal detection in the no-sample negative control reaction is PCR contamination. We recommend repeating the RT-PCR with fresh reagents and freshly decontaminated pipettors. Be sure to set up the RT-PCR in an area isolated from areas used for RNA isolation and PCR product analysis

QUALITY CONTROL

Relevant kit components are tested in the following nuclease assays.

RNase Activity:

A sample is incubated with labeled RNA, followed by PAGE analysis.

Nonspecific Endonuclease Activity:

A sample is incubated with supercoiled plasmid DNA and analyzed by agarose gel electrophoresis.

Exonuclease Activity:

A sample is incubated with labeled double-stranded DNA, followed by PAGE analysis.

OTHER INFORMATION

Material Safety Data Sheets:

Material Safety Data Sheets (MSDSs) for any chemical product supplied by Applied Biosystems or Ambion are available 24 hours a day. At www.appliedbiosystems.com, select Support, then MSDS. Search by chemical name, product name, product part number, or MSDS part number. Right-click to print or download the MSDS of interest. At www.ambion.com, go to the web catalog page for the product of interest. Select MSDS, then right-click to print or download. Or, e-mail (MSDS_Inquiry_CCRM@appliedbiosystems.com), telephone (650-554-2756; USA), or fax (650-554-2252; USA) your request, specifying the catalog or part number(s) and the name of the product(s). We will e-mail the associated MSDSs unless you request fax or postal delivery. Requests for postal delivery require 1-2 weeks for processing.

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