AZscript™ Reverse Transcriptase

AZscript Reverse Transcriptase (RT) is an RNA-dependent DNA polymerase used to synthesize complementary DNA (cDNA) from an RNA template.

While originating from Moloney Murine Leukemia Virus (M-MLV), AZscript RT has been engineered for increased thermostability and reduced RNase H activity.

In cDNA synthesis a higher reaction temperature can reduce RNA secondary structures and improve efficiency and specificity of the reaction. Furthermore, a reduction in RNase H activity increases performance of the cDNA synthesis, especially for longer transcripts.

AZscript RT shows excellent performance and sensitivity in RT-qPCR (Fig 1).

Increased thermostability

Reduced RNase H activity

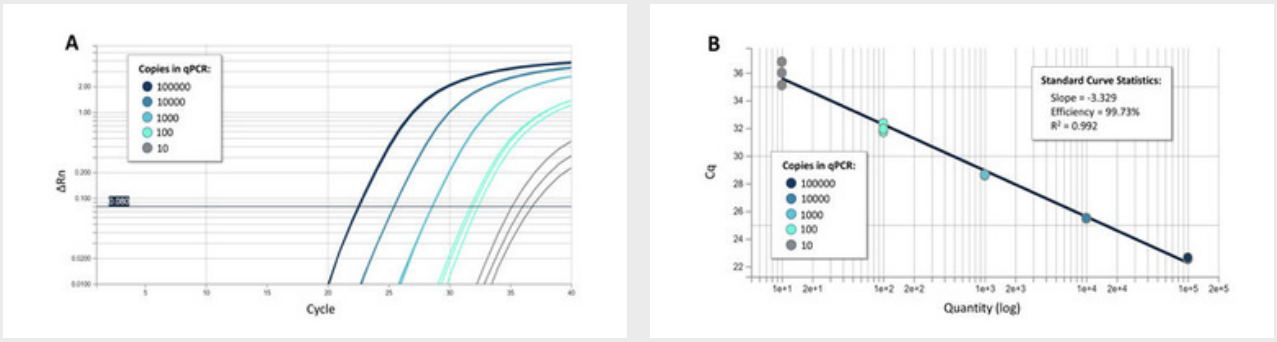

Fig 1. AZscript RT performance in RT-qPCR

AZscript shows excellent performance and sensitivity in RT-qPCR. A serial dilution of MS2 Armored RNA (Asuragen, 52000) was heat lysed (75°C, 5min) and used as template for cDNA synthesis using gene-specific primers, according to manufacturer's instructions. A volume equivalent to 2 µl of the cDNA was amplified in a 25 µl reaction using TaqPath™ qPCR Master Mix, CG. Data showing amplification plots (A) and standard curve (B) for the dilution series corresponding to 100 000 down to 10 copies of RNA, in triplicate measurements.

Specifications

Source	Recombinantly produced in *E. coli*.
Size	79.5 kDa
Temperature optimum (>80%)	38 - 47°C
Inactivation	70°C, 15 minutes

Storage and stability

The enzyme is stable at -20°C for up to 12 months in the supplied storage buffer. Additional data on stability of AZscript RT is available on request.

Quality control

dsDNA endonuclease activity	200 U AZscript RT was incubated with a supercoiled plasmid (1 µg) for 4 hours at 37°C. Agarose gel electrophoresis did not reveal any transformation of closed circular DNA to nicked DNA.
ssDNA endonuclease activity	200 U AZscript RT was incubated with M13 ssDNA (0.5 µg) for 4 hours at 37°C. Agarose gel electrophoresis did not reveal any visible signs of ssDNA degradation.
Exonuclease activity	200 U AZscript RT was incubated with either 3H-dATP labelled ds or ssDNA (0.5 µg, 500 bp) for 4 hours at 37°C. Acid soluble radioactivity from labelled DNA was not significantly over blank test for either substrate.
RNase activity	200 U AZscript RT was incubated with a 2 kb RNA transcript (1 µg) for 4 hours at 37°C. Agarose gel electrophoresis did not reveal any visible signs of RNA degradation.

Ordering information

	Article no.	Pack Size*	Concentration
AZscript™ RT	72100-201	10 000 U	200 U/µl
	72100-100	Custom	Custom

*One unit is defined as the amount of enzyme needed to incorporate 1 nmol of dTTP into acid-precipitable material in 10 min at 37°C using poly(A)●oligo(dT)15 as template-primer. The enzyme is assayed in 37.5 mM Tris-HCl pH 8.3 @ 25°C, 40 mM KCl, 6 mM $MgCl_2$, 0.75 mg/ml Poly(A), 0.03 mM oligo(dT)15, 10 mM DTT, 0.5 mM dTTP, 0.025 mCi/ml 3H-dTTP, 0.1 mg/ml BSA, 0.02% Triton X-100.

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Quality

ArcticZymes is dedicated to the quality of its products and is certified according to ISO 13485. ArcticZymes offers the convenience of providing standard bulk enzymes as off the shelf products. In addition, ArcticZymes offers enzymes in customized formats. For additional information, please contact us.

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Additional Information

We are pleased to provide additional data and information relating to AZscript reverse transcriptase on request. For more information about our enzymes and services, please visit our website www.arcticzymes.com.