

PrimeScript™ RT reagent Kit with gDNA Eraser (Cat.# RR047A)

Application: Comparison of cDNA yield from PrimeScript RT reagent Kit with and without genomic DNA elimination

The PrimeScript RT reagent Kit with gDNA Eraser (Cat.# RR047A) is a reverse transcription kit for real-time RT-PCR that includes a genomic DNA elimination step. The kit is designed to avoid loss of RNA.

Methods:

1. Reverse Transcription

Reagent:	PrimeScript RT Reagent Kit with gDNA Eraser (Perfect Real Time) (Cat.# RR047A) or PrimeScript RT Reagent Kit (Perfect Real Time) (Cat.# RR037A)
Template:	Mouse liver total RNA (2 pg–2 µg). For Cat.# RR047, 200 ng of mouse genomic DNA was added to each total RNA.
Reaction volume:	20 µl
Primer:	RT Primer Mix
Reaction conditions:	Standard conditions recommended in the User Manual of each kit

2. Real Time PCR

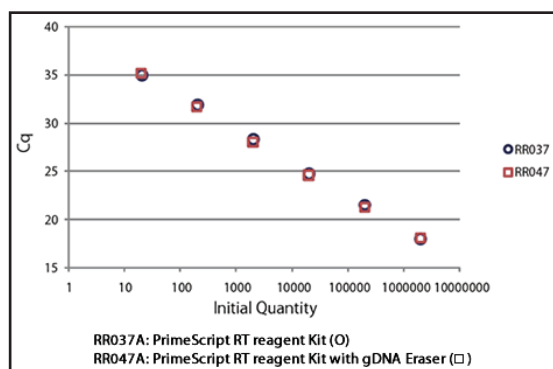
Reagent:	SYBR® <i>Premix Ex Taq</i> ™ II (Perfect Real Time) (Cat.# RR081A)*
Template:	Reverse transcription product, 2 µl each
Reaction volume:	25 µl
Target gene:	Mouse <i>Rsp18</i>
Reaction conditions:	Standard protocol of the Thermal Cycler Dice Real Time System**

*Cat.# RR081A has been discontinued. SYBR® *Premix Ex Taq*™ II (Tli RNase H Plus) (Cat.# RR820A) would be a suitable substitution.

**Not available in all geographic locations. Check for availability in your region.

Results:

The standard curve was plotted for each reverse transcription kit.



Reagent	R ²	Amplification Efficiency (%)	Standard Curve
RR037A	1.000	95.9	$Y = -3.425 \times \text{LOG}(X) + 43.05$
RR047A	0.999	95.7	$Y = -3.429 \times \text{LOG}(X) + 42.94$

Conclusions:

Amplification efficiency observed using Cat.# RR037A and Cat.# RR047A was nearly identical using RNA template amounts ranging from 2 pg–2 µg. The gDNA removal step does not lead to degradation or loss of template RNA.