

AllScript II Green One-Step qRT-PCR SuperMix



Cat. No. ABTGMBR311-100

Storage: Store components in the dark at -20°C for one year

Description

AllScript II Green One-Step qRT-PCR SuperMix is a one-step RT-PCR kit , Which cDNA synthesis and PCR are performed in a single tube using gene-specific primers (GSP) and either total RNA or mRNA.

Component	100 rxns
AllScript II Green One-Step RT/RI Enzyme Mix	40 µl
2×AllScript Green qPCR SuperMix	1 ml
RNase-free Water	1 ml
* Passive Reference Dye/PCR Enhancer (50×)	40 µl

* Passive Reference Dye is optional. Please refer to the comparison as below.

Corresponding instrument

- Passive Reference Dye I (50×):

ABI Prism 7000/7700/7900, Eppendorf, ABI Step One Plus.

- Passive Reference Dye II (50×):

ABI Prism 7500, ABI Prism 7500 Fast, ABI Q6, ABI Quant Studio 6/7 Flex, ABI ViiA7, Stratagene Mx3000/Mx3005P, Corbett rotor gene 3000.

- No Passive Reference Dye:

Roche LightCycler480, Roche Light Cycler 96, MJ Research Chromo4, Opticon(II), Takara TP800, Bio-Rad iCycler iQ, iQ5, Bio-Rad CFX96, Bio-Rad C1000 Thermal Cycler, Thermo Pikoreal 96, Corbett Rotor Gene 6000, Corbett Rotor Gene G, Corbett Rotor Gene Q.

Reaction components

Component	Volume	Final Concentration
RNA Template	1pg~1µg	as required
Forward GSP (10 µM)	0.4 µl	0.2 µM
Reverse GSP (10 µM)	0.4 µl	0.2 µM
2×AllScript Green qPCR SuperMix	10 µl	1×
AllScript II Green One-Step RT/RI Enzyme Mix	0.4 µl	-
* Passive Reference Dye or PCR Enhancer(50×)	0.4 µl	1×
R Nase-free Water	Variable	-
Total volume	20 µl	-

* Passive Reference Dye is optional.

Thermal cycling conditions (three-step)	
50°C	5 min
94°C	30 sec
94°C	5 sec
50-60°C	15 sec *
72°C	10 sec *
Dissociation Stage	

40-45 cycles

Thermal cycling conditions (two-step)	
50°C	5 min
94°C	30 sec
94°C	5 sec
60°C	30 sec *
Dissociation Stage	

40-45 cycles

Fluorescent signals can be collected during the annealing or extension stage. For ABI qPCR instrument, suggest to use the following signal collecting time :

- * For ABI Prism 7700/7900, the time to 30 sec.
- * For ABI Prism 7000/7300, the time to 31 sec.
- * For ABI Prism 7500, the time to 34 sec.
- * For ABI ViiA7, the time is at least 19 sec.

Two-step qPCR is more suitable for higher specificity assay.

Three-step qPCR is more suitable for higher sensitivity assay.