



## ampliTune® qPCR EvaGreen® Mix (5X) with ROX

Cat. No.	Pack Size	Conc. (MgCl <sub>2</sub> )
11025	0.1 ml SAMPLE	12.5 mM
11250	1 ml	12.5 mM

### Description

ampliTune® qPCR EvaGreen® Mix is a pre-formulated master mix for real-time qPCR assays. The improved composition of the mix makes it usable on standard and fast qPCR platforms irrespectively of ROX-requirements. Coupled with user-supplied primer sets and template, ampliTune® qPCR EvaGreen® Mix provides exceptional accuracy and high reproducibility during target amplification.

A 12 min incubation step at 95°C activates the polymerase and prevents primer dimer formation and non-specific amplification during qPCR.

### Advantages

- ✓ Specific real-time PCR and reproducible results
- ✓ Increased sensitivity for low copy number targets
- ✓ Superior performance with long (≤500 bp) and GC-rich templates
- ✓ UNG treatment capability to prevent carryover contamination from previous runs<sup>1</sup>
- ✓ Stable at room temperature for up to 1 month
- ✓ Easy control during pipetting due to visualization dye

<sup>1</sup>UNG is not included and must be purchased separately.

### Storage & Shipping

Store at -20°C, shipping at room temperature. Store in dark. Shipping and temporary storage for up to 1 month at room temperature has no detrimental effects on the quality of ampliTune® qPCR EvaGreen® Mix.

### Reagent composition

ampliTune® DNA polymerase is supplied in a 5X qPCR buffer, dNTPs including dUTP, 12.5 mM MgCl<sub>2</sub> (1x PCR solution – 2.5 mM MgCl<sub>2</sub>), EvaGreen® dye, internal reference based on ROX, GC-enhancer and blue visualization dye.

### Recommended qPCR reaction mix

Component	Volume	Final conc.
ampliTune® qPCR EvaGreen® Mix (5X)	4 µl	1X
Primer Forward (10 pmol/µl)	0.2-0.4 µl	0.1-0.2 µM
Primer Reverse (10 pmol/µl)	0.2-0.4 µl	0.1-0.2 µM
DNA template	1-5 µl	variable <sup>2</sup>
H <sub>2</sub> O PCR grade	up to 20 µl	
<b>Total</b>	<b>20 µl</b>	

<sup>2</sup>cDNA 0.1 pg/µl – 10 ng/µl; gDNA 10 pg/µl – 4 ng/µl

### Recommended qPCR cycles

Cycle step	Temp.	Time	Cycles
Optional UNG treatment	50 °C	2 min	1
Initial activation	95 °C	12 min	1
Denaturation	95 °C	15 s	} 40
Annealing	60-65 °C	20-30 s <sup>3</sup>	
Elongation	72 °C	20-30 s <sup>3</sup>	

<sup>3</sup>Use 20 sec for annealing and elongation for templates shorter than 150 bp.

### Product use limitation

This product is developed and supplied for research purposes and *in vitro* use only. Please refer to [www.selectabiotech.com](http://www.selectabiotech.com) for Material Safety Data Sheet of the product.

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ampliTune® is a registered trademark of Selecta Biotech SE

### CERTIFICATE OF ANALYSIS

#### Endodeoxyribonuclease activity

No contaminating endonuclease activity was observed upon incubation of 10 µl of ampliTune® qPCR EvaGreen® Mix (5X) with 1 µg of pUC19 DNA in 50 µl of reaction mixture for 4 hours at 37°C.

#### Exodeoxyribonuclease activity

No contaminating exonuclease activity was detected after incubation of 10 µl of ampliTune® qPCR EvaGreen® Mix (5X) with 1 µg of digested DNA in 50 µl of reaction mixture for 4 hours at 37°C.

#### Ribonuclease activity

No contaminating RNase activity was detected after incubation of 1 µg of RNA transcript with 10 µl of ampliTune® qPCR EvaGreen® Mix (5X) for 4 hours at 37°C.

#### Functional assay

ampliTune® qPCR EvaGreen® Mix (5X) was tested for amplification of 75, 196 and 343 bp DNA fragments.

Quality authorized by: Mgr. Petra Mazancova, PhD. 



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