



	Related products	Catalog No.
Catalog No.: R-EMX-01/02/03/04	RT All-in-One Master Mix	R-G208
Product Name: EvaGreen qPCR PreMix	Direct RT-qPCR Lysis Kit	R-G915
Size: 1.25ml (125rxns of 20µl)	RT Genomic DNA Removal Kit	R-G488
Storage: -20°C and avoid light		

Description: EvaGreen qPCR PreMix is designed for quantitative/real-time analysis. The components of EvaGreen qPCR PreMix has been developed for superb performance in sensitivity, signal-to-noise ratio, and complete elimination of primer dimers. The chemically modified Hot-Start Taq polymerase, included in our master mix, significantly reduces non-specific PCR amplification observed with regular Taq polymerase. Every lot is tested on different real-time PCR machines listed below.

- Special Features:** ★ Instrument-specific, pre-optimized real-time PCR reagents
- ★ Validated application
 - ★ Reproducible results
 - ★ Best Price

Application: EvaGreen qPCR PreMix is ideally suited for:

- Gene expression analysis
- Microarray validation
- Viral load determination

Kit Contents: EvaGreen qPCR PreMix is a 2X mix of dNTPs, Hotstart Taq polymerase, MgCl₂, fluorescent detection dye, reference dye, and proprietary buffer components.

Note: After each experiment, the leftover thawed mix can be stored at 4°C if it is to be used within the next 3 months. Avoid repeated freeze-thaw cycles to retain maximum performance. **EvaGreen qPCR PreMix** is stable for 1 year from the date of shipping when stored and handled properly.

**Guideline for your particular instrument model:**

Due to variations in qPCR instruments, we offer different **EvaGreen qPCR Master Mix** formulations, optimized for different machines. Please use the following table as a guideline for the selection of qPCR Master Mix appropriate for your particular instrument model.

Cat No.	Product Name	Recommended Machines
R-EMX-01	EvaGreen qPCR PreMix-ROX	ABI®7000 series (except 7500), StepOne™, PRISM® 7000 series, and Gene Amp 5700
R-EMX-02	EvaGreen qPCR PreMix-Low ROX	Any qPCR machine that needs low ROX dye; such as, ABI® 7500, Stratagene® Mx3000, Mx3005, Mx4000 and Analytikjena's qTower
R-EMX-03	EvaGreen qPCR PreMix-iCycler	BioRad® iCycler®, iQ™5 and MyiQ™ Thermal Cycler
R-EMX-04	EvaGreen qPCR PreMix-No Dye	Any qPCR machine that does not need reference dye; such as Eppendorf® Roche LightCycler®, MJ Research, Qiagen, Corbett, Bioer Technology and DNA-Technology

General Protocol**Reaction Setup:**

1. Thaw **EvaGreen qPCR PreMix**, template DNA, primers and RNase-free water on ice. Mix each solution well.
2. Prepare a reaction Master Mix using the following:

Components	Volume 20µl	Volume 25µl	Volume 50µl	Final Concentration
EvaGreen qPCR PreMix	10µl	12.5µl	25µl	1X
Primer A (10µM*)	0.2-1.0µl	0.25-1.25µl	0.5-2.5µl	100-500nM
Primer B (10µM*)	0.2-1.0µl	0.25-1.25µl	0.5-2.5µl	100-500nM
Sterile Water	Variable	Variable	Variable	
Template DNA	Variable	Variable	Variable	= or <500ng/reaction
Total Volume	20µl	25µl	50µl	
* Suggested concentration.				

3. Perform qPCR reactions using the following cycling program.

Step	Temperature	Duration-Standard	Duration-Fast	Cycles
Enzyme Activation	95°C	10min	10min	Hold
Denature	95°C	15sec	3sec	40
Anneal/extend	60°C	60sec	30sec	
Melting Curve	According to the instrument guidelines			

Recommendations for Optimal Results:

- Aliquot reagents to avoid contamination and to avoid repeated freeze-thaw cycles.
- **EvaGreen qPCR PreMix** components are light sensitive; avoid exposure to light.
- Start PCR as soon as the reaction mixture is prepared and always keep the reaction mixture chilled in an ice box prior to PCR reactions.

This product is for research use only.