

Product Description

AcuGenix™ Superstart Lyo-Ready All-in-One qPCR Master Mix with UNG is a dedicated reagent developed for lyophilization processes, designed for qualitative and quantitative real-time PCR detection using probe-based chemistry. It contains a novel hot-start enzyme, Superstart Taq plus (DG), in which Taq polymerase activity is blocked at room temperature. This effectively suppresses non-specific amplification caused by non-specific primer annealing or primer-dimers under low-temperature conditions, thereby improving amplification specificity. The reagent employs an optimized qPCR-specific buffer and a UNG/dUTP carryover prevention system, enabling rapid hot-start activation and significantly enhancing the amplification efficiency and detection sensitivity of qPCR reactions. It yields a good standard curve over a wide quantitative range, allowing accurate quantification, and effectively prevents false-positive amplification caused by PCR product carryover or aerosol contamination. The reagent is compatible with most fluorescence quantitative PCR instruments from manufacturers such as Applied Biosystems, Eppendorf, Bio-Rad, and Roche. After being freeze-dried, this product maintains good stability in terms of its freeze-drying morphology.

Components

1. 5×Superstart Premix plus-UNG (Mg²⁺ free) (DG)
2. 250 mM MgCl₂
3. 4×AcuGenix™ Lyophilization Protectant-9 (optional)

Storage

Store at -20±5℃. Vortex before use. Avoid freeze-thaw cycles.

Prepare Reaction Mix

Composition	Volume per Reaction	Volume per Reaction	Final Concentration
5×Superstart Premix plus-UNG (Mg ²⁺ free) (DG)	5 µL	10 µL	1×
250 mM MgCl ₂	0.45 µL	0.9 µL	4.5 mM
4×AcuGenix™ Lyophilization Protectant-9 ¹	6.25 µL	12.5 µL	1×
25×Primer-Probe Mix ²	1 µL	2 µL	1×
Template DNA ³	---	---	---
ddH ₂ O	To 25 µL	To 50 µL	---

1. This system is lyophilizable. When customers use this system without the need for lyophilization, the 4×AcuGenix™ Lyophilization Protectant-9 may be added optionally. If lyophilized products are required, the 4×AcuGenix™ Lyophilization Protectant-9 must be added during product performance verification at the liquid reagent stage to ensure consistency in composition and performance with the lyophilized system.

2. Usually, a final primer concentration of 0.2 µM can yield good results, when the reaction performance is poor, the primer concentration can be adjusted within the range of 0.2-1 µM. Typically, probe optimization is performed in the range of 0.1-0.3 µM. Concentration gradient experiments can be conducted to find the optimal combination of primers and probes.

3. The copy number of target genes varies in different types of templates, when necessary, dilute gradients to determine the optimal amount of template addition.

Reaction Programe

Standard PCR Program			
Procedure	Temp	Time	Cycle
Digestion	50℃	2 min	1
Hot Start	95℃	1-5 min	1
Denaturation	95℃	10-20 s	40-50
Annealing and Extension	56-64℃	20-60 s	

Prepare Reaction Mix for lyophilization

Components	25 µL System
5×Superstart Premix plus-UNG (Mg ²⁺ free) (DG)	5 µL
250 mM MgCl ₂	0.45 µL
4×AcuGenix™ Lyophilization Protectant-9	6.25 µL
25×Primer-Probe Mix	1 µL
ddH ₂ O	To 18-20 µL

*For other lyophilization system formulations, please inquire separately.

Lyophilization conditions

Procedure	Temperature	Time	status	Pressure
Pre-freezing	4℃	30 min	Hold	1 atm
	-50℃	60 min	Cooling	
	-50℃	180 min	Hold	
Primary drying	-30℃	60 min	Heating	Ultimate vacuum
	-30℃	720 min	Hold	
Secondary drying	25℃	60 min	Heating	Ultimate vacuum
	25℃	300 min	Hold	

1. This freeze-drying process is an in-situ freeze-drying process for a 25 µL system. If need to perform freeze-drying of freeze-dried beads or other in-situ systems, please inquire separately.
2. The above freeze-drying process is for reference only. Different product types and the use of different freeze dryers may result in different parameters, so adjustments can be made based on actual usage.

Quality Control

1. Functional testing: qPCR sensitivity, specificity, repeatability.
2. No exogenous nuclease activity, no exogenous endonuclease/exonuclease contamination.

Notes

3. The amplification rate provided by DNA polymerase contained in this product is not less than 1 kb/10 s. Different fast PCR instruments have different temperature control modes, rates, and thermal conductivity efficiencies. It is recommended that specific optimization for each instrument's most suitable reaction conditions be carried out along with their respective fast PCR instruments.
4. This reagent has higher specificity and significantly improves fluorescence quantitative PCR limit detection sensitivity, making it suitable as a high-sensitivity fluorescence quantitative PCR test agent for improving curve uniformity and fluorescent values at extremely low template concentrations.
5. For primers with lower annealing temperatures or fragments over 200 bp, it is suggested that three-step amplification be used.
6. Different target genes vary in their utilization efficiency of dUTP and sensitivity to UNG. Therefore, if the use of a UNG system results in reduced detection sensitivity, the reaction system should be adjusted and optimized. For technical support, please contact our company.
7. Use dedicated areas and pipettes before and after amplification, wear gloves during operation, and regularly replace them. Do not open reaction tubes after PCR completion to minimize sample contamination from PCR products.