

Multiplex PCR MasterMix (UNG)

Project number: M665822

Storage condition: -20°C, try to avoid repeated freezing and thawing

Product content

Component	M665822-5×1ml
2×Multiplex PCR MasterMix (UNG)	5 x 1ml
ddH ₂ O	5 x 1ml

Product Introduction

2×Multiplex PCR MasterMix (UNG) is a PCR premix system consisting of GoldStar Taq DNA Polymerase, Mg²⁺, dNTPs (including dUTP), UNG enzyme and PCR stabilizer. This product eliminates the need for complicated optimization of PCR reaction conditions and allows for easy multiplexing of PCR reactions by simple mapping of conditions.

The GoldStar Taq DNA Polymerase contained in this product is a chemically modified hot-start enzyme that effectively reduces non-specific amplification due to primer mismatches at the beginning of the PCR reaction. The unique buffer system allows all primers of the multiplex PCR reaction to be extended efficiently without additional optimization. The MasterMix also includes a GC Enhancer, which helps to achieve efficient amplification of "difficult" templates (e.g., those with high GC content). False positives due to contamination of amplification products are greatly reduced by the use of the dUTP-UNG Anti-Contamination System, which effectively removes residual contamination of PCR products. ung enzyme is inactivated during the pre-denaturation step of the PCR cycle, so that it does not interfere with the formation of new PCR products containing dU bases.

Multiplex PCR MasterMix (UNG) effectively prevents residual contamination of PCR products and is suitable for contamination-proof multiplex PCR reactions such as microsatellite analysis, genotyping and SNP detection.

quality control

No exogenous nuclease activity was examined; no host residual DNA was detected by PCR method; and the amplification performance was not significantly altered by storage at 2-8° C for 3 days.

Usage

The following examples are conventional PCR reaction systems and reaction conditions, which should be improved and optimized according to the template, primer structure and target fragment size in actual operation.

1. PCR reaction system:

reagents	50 µl reaction system	final concentration
2×Multiplex PCR MasterMix (UNG)	25 µl	1×
Primer Mix, 10µM each	1 µl	0.2 µM
Template DNA	appropriate amount	
ddH ₂ O	up to 50 µl	

Note: When primer design, try to minimize the difference between the T_m of each primer, and the difference should be controlled within 5°C as much as possible.

Please use the final concentration of 0.05–0.2 μ M as the reference for setting the range of each primer concentration. If the amplification efficiency is not high, the concentration of primers can be increased; when non-specific amplification occurs, the concentration of primers can be decreased, thus optimizing the reaction system.

2. PCR reaction conditions:

Steps	Temperature	time
UNG enzyme digestion	50° C	2–10min
Pre denaturation	95° C	10min
denaturation	95° C	30s
annealing	55–65° C	30s
Extend	72° C	60s/kb
Final extension	72° C	5min

} 30–40 cycles

Note: 1) In general, the annealing temperature in the experiment is 5° C lower than the melting temperature T_m of the amplification primer, and when the ideal amplification efficiency cannot be obtained, the annealing temperature is appropriately lowered; when a non-specific reaction occurs, the annealing temperature is raised, thus optimizing the reaction conditions.

2) The extension time should be set according to the size of the fragment being amplified. The amplification efficiency of GoldStar Taq DNA Polymerase included in this product is 1kb/min.

3) The number of cycles can be set according to the downstream application of the amplification product. If the number of cycles is too low, the amplification is insufficient; if the number of cycles is too high, the chance of mismatch will increase and the non-specific background will be serious. Therefore, the number of cycles should be minimized under the premise of ensuring the product yield.

3. Result detection: This product does not contain dyes, after the reaction is finished, take 5 μ l of the reaction product and add the appropriate amount of sampling buffer for electrophoresis to detect the results.