AcuGenix™ Super-Fidelity Master Mix V01

Product Description

AcuGenix™ Super-Fidelity Master Mix is a ready-to-use 2×high-fidelity PCR premix containing a genetically engineered hot-start high-fidelity DNA polymerase. The enzyme has extremely high DNA affinity and processivity and shows excellent compatibility with complex templates and partially degraded templates.Its fidelity is approximately 154-fold higher than that of Taq DNA polymerase. Unique extension factors and specificity-enhancing factors are included to greatly improve long-fragment amplification capability, amplification specificity, and amplification yield.

When simple templates such as λDNA or plasmids are used, 2×Super-Fidelity Master Mix can efficiently amplify fragments up to 30 kb. When complex templates such as genomic DNA are used, it can amplify fragments up to 20 kb. In addition, 2×Super-Fidelity Master Mix shows good resistance to PCR inhibitors and can be used for direct PCR of bacteria, fungi, plant tissues, or whole-blood samples.

2×Super-Fidelity Master Mix contains hot-start high-fidelity DNA polymerase, dNTPs, and an optimized buffer system. Amplification can be performed by adding only primers and template, reducing pipetting steps and improving assay throughput and result reproducibility. A version containing electrophoresis tracking dye is also available, allowing direct gel loading after PCR. This product is suitable for applications such as gene-cluster cloning and ultra-long-fragment assembly.

Product Information

Product Name	Cat. No.	Volume
AcuGenix™ Super-Fidelity Master Mix (Dye Plus)	BR3M121-79	0.1 mL
	BR3M121-71	1 mL
	BR3M121-72	5×1 mL
	BR3M121-74	15×1 mL
AcuGenix™ Super-Fidelity Master Mix	BR3M121-09	0.1 mL
	BR3M121-01	1 mL
	BR3M121-02	5×1 mL
	BR3M121-04	15×1 mL

Storage

Store at -20±5℃.

Notes

1. This product is intended for scientific research purposes only.
2. Do not use dUTP or primers and templates containing uracil.
3. The DNA polymerase is modified with a monoclonal antibody. The reaction system can be prepared at room temperature, but it is recommended to keep the reagent on ice before use and return it to the freezer promptly after use.
4. When amplifying fragments >10 kb, primer lengths >26 nt are recommended. Primer design should follow general principles.

Protocol

1. PCR Reaction Setup

1.1 Thaw all components at room temperature or on ice, mix thoroughly, and briefly centrifuge. Prepare the reaction mix according to Table 1.

Table 1. PCR Reaction Setup

Component	Volume (μL)	Remarks
Template [a]	X	
AcuGenix™ Super-Fidelity Master Mix	25	1×
Primer 1 (10 μM)	0.5-2	0.1-0.4 μM [b]
Primer 2 (10 μM)	0.5-2	0.1-0.4 μM
ddH_2O	Up to 50	

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Note a: The optimal reaction concentration varies by template. Table 2 lists recommended template amounts for a 50 μL reaction system.

Note b: In general, a final primer concentration of 0.2 μM is sufficient. If non-specific amplification occurs, reducing the final primer concentration to 0.1 μM can improve specificity.

Table 2. Recommended PCR Template Amounts

Template Type	Recommended Template Amount
Genomic DNA	50-400 ng
Plasmid or viral DNA	10 pg-30 ng
cDNA	1-5 μL (not exceeding 1/10 of the total PCR reaction volume)
Crude template [c]	1-2 μL recommended

Note c: For whole-blood samples, adding 1%-2% whole blood to the PCR reaction system is recommended. The initial denaturation time in the PCR program should be adjusted to 5 min. After PCR, centrifuge at 4000 rpm for 1 min to pellet blood-cell debris, and use the supernatant for downstream analysis. For fungal, bacterial, animal-tissue, or plant-tissue samples, cell lysis with lysis buffer before PCR is recommended.

1.2 Gently pipette to mix and briefly centrifuge to collect the reaction solution at the bottom of the tube.

2. PCR Cycling Program

2.1 Set the PCR cycling program on the thermal cycler according to Table 3.

Table 3. PCR Cycling Program

Step	Temperature	Time	Cycles
Heated lid	105℃	/	/
Initial denaturation	98℃	30 s [a]	1 cycle
Denaturation	98℃	15 s	25-35 cycles
Annealing	56-72℃ [b]	15 s	
Extension	72℃ [c]	5-20 s/kb	
Final extension	72℃	3-5 min	1 cycle
Hold	4℃	∞	1 cycle

Note a: For most templates, set initial denaturation to 30 s. If the template has high GC content, extend the initial denaturation to 1 min.

Note b: Set the annealing temperature according to the primer Tm. If the primer Tm is relatively high, the annealing step can be omitted and a two-step PCR program can be used. When using a two-step program, the annealing/extension temperature can be optimized between 65℃ and 68℃. If needed, establish a temperature gradient to identify the optimal primer-template binding temperature. The annealing temperature directly determines amplification specificity; if specificity is poor, increase the annealing temperature appropriately.

Note c: For target fragments of 1-4 kb, set the extension time to 1-5 s/kb; for 4-8 kb, use 10 s/kb; for 8-12 kb, use 10-15 s/kb; for targets >12 kb, 20 s/kb is recommended. If the amplification yield is low, increase the primer concentration or extend the extension time appropriately.