# Lolina A/S

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## **Product Specification**

Product name	2 × Lolina® HotStart PCR Genotyping Master Mix (With Dye)		
Cat.No.	NaM201007-2		
Size	1 mL/5×1 mL/50×1 mL/100×1 mL		
Storage and shipping	The product is shipped with dryice and can be stored at -20°C for 2 year.		
Application	For mouse genotyping mainly		
QC	<ul> <li>Exonuclease residue detection: 20 μL of this product and 0.6 μg λ DNA-Hind III were incubated for 4 h at 37°C. There was no change in the electrophoresis band of DNA.</li> <li>Endonuclease residue detection: 20 μL of this product and 1 μg of λDNA were incubated at 37°C for 4 hours. There was no change in the DNA electrophoresis band.</li> <li>Detection of Escherichia coli residual DNA: Add 25 μL of this product to a 50 μL system, and use sterile ddH<sub>2</sub>O as a template to amplify the E.coil 16s rDNA gene. After 30 cycles, the amplified products were subjected to 1% agarose gel electrophoresis and EB staining, and no amplified bands were found.</li> </ul>		

### **Product description**

 $2 \times \text{Lolina}$  HotStart PCR Genotyping Master Mix (With Dye) is a ready-to-use PCR premix solution, containing Lolina® HotStart Taq DNA Polymerase, dNTPs and an optimized buffer system. Just add primers and templates for amplification, greatly simplifying the experimental steps and enabling high-throughput operation and improve the reproducibility of experimental results. Lolina® HotStart Taq DNA Polymerase is a thermostable Taq DNA Polymerase modified with a ligand that modulates DNA polymerase activity as a function of temperature. Enzyme activity is completely blocked at room temperature and is released after heating to 95°C. Lolina<sup>®</sup> HotStart DNA Polymerase requires only 2-3 minutes to activate and is compatible with existing PCR protocols. This product prevents non-specific amplification during sample preparation and reaction heating stages, and can effectively perform genotyping experiments.

#### Instructions

#### 1. Recommended PCR Reaction System (50 $\mu$ L)

Components	Volume µL	Final concentration
2 × Lolina <sup>®</sup> HotStart PCR Genotyping Master Mix (With Dye)	25	1 ×
Template	х	-
Forward Primer (10 µmol/L)	2	0.4 μmol/L
Reverse Primer (10 µmol/L)	2	0.4 μmol/L
ddH <sub>2</sub> 0	Up to 50	-

[Note]:

The optimal reaction concentration for different templates is different. The following table is the recommended template usage for a 50  $\mu$ L reaction system, which is for reference only.

Components	Volume μL
Genomic DNA	50 ng-100 ng
Plasmid DNA	100 pg-20 ng
cDNA	1-5 $\mu$ L (no more than 1/10 of the reaction system)

#### 2. Reaction program

Cycle step	Temp.	Time	Cycles
Initial denaturation	95 °C	5 min	1
Denaturation	95 °C	30 sec	
Annealing	50-60 °C	30 sec	35
Extension	72 °C	30 sec	
Final extension	72 °C	10 min	1

[Note]:

1) Annealing temperature and time: The recommended annealing temperature is  $50-60^{\circ}$ C. The recommended annealing time is 30 sec, which can be adjusted within 20-30 sec. As needed, a temperature gradient can be set up to find the optimal temperature and time for index annealing.

2) Extension temperature and time: The recommended temperature is 72°C. The recommended time is 30-60 sec/kb.

3) Amplification product: Please store the PCR amplification product at -20 $^{\circ}$ C to prevent DNA degradation.

### Notes

1. This product is for research use only.

2. Please operate with lab coats and disposable gloves for your safety.