



KlearKall Master mix

(For research use only. Not for use in diagnostic procedures.)

****Please ensure that the kit is stored at -20°C****

Introduction

KlearKall Master mix is a convenient 2X mix for TaqMan probe-based genotyping, and contains all required components except sample DNA, water, and a probe-based assay.

The KlearKall Master mix contains:

- KlearTaq™ Hot Start DNA polymerase, a purified DNA polymerase. This is inactive at room temperature, so reactions can be set up on the bench top. The enzyme is activated during thermal cycling.
- Optimized components including buffer and dNTPs for consistent, reliable genotypes.
- ROX to use as passive internal reference dye for precise data analysis

KlearKall Master mix is compatible with most qPCR instruments and FRET-capable plate readers.

Key features

- Specifically formulated for endpoint fluorescent detection for probe-based genotyping
- Discrete clusters and high call rates for accurate and reproducible allelic discrimination
- Universal thermal cycling conditions for consistent results
- Bench top stability for ease and flexibility of experiment set up.

Storage conditions

KlearKall Master mix can be safely stored for at least one week at 4°C, one year at -20°C or indefinitely at -80°C. If the KlearKall Master mix is divided into aliquots, it is recommended that the tubes used are light-protective. Frequent freezing and thawing should be avoided for the optimal performance.

Customer requirements

1. Nuclease-free water
2. Probe-based assay e.g. TaqMan
3. Template DNA.

General guidelines

1. The KlearTaq enzyme in KlearKall Master mix requires a 15 minute initial activation stage (95°C).
2. dTTP is used in our KlearKall Master mix as we have found that it improves reaction sensitivity and efficiency when compared to mixes containing dUTP.
3. The purchase of this kit to perform the 5' nuclease assay does not convey a license to any patents that may be in existence for this technology.

Reaction set-up

KlearKall Master mix is supplied at 2X concentration and should be diluted to 1X concentration in the final reaction. A typical reaction setup should include KlearKall Master mix, a probe-based assay, and template DNA. Recommended total reaction volumes are:

- 10 - 25 μL for 96-well plates
- 3 - 5 μL for 384-well plates
- 1 - 3 μL for 1536-well plates.

Protocol

1. Completely thaw all of the reaction components and briefly vortex before use. Briefly spin the tubes in a microcentrifuge to ensure that the material is collected at the bottom of the tube.
2. Combine all components of the PCR reaction in the wells of a reaction plate.
3. Briefly spin the reaction plate in a microcentrifuge to ensure that the material is collected at the bottom of the plate.
4. Place the reactions in a thermal cycler / qPCR instrument and perform the thermal cycle. An example thermal cycle program is detailed in Table 1.

Table 1: An example of a qPCR thermal cycle program

Step	Temperature	Time	Number of cycles
1	95	15 min	1 cycle
2	95	15 sec	40 cycles
	60	60 sec	

Ordering information

Product code	Product name	Description
KBS-1002-001	2X KlearKall 100 Std ROX	2.5 mL, contains ROX
KBS-1002-003	2X KlearKall 1000 Std ROX	25 mL, contains ROX
KBS-1002-007	2X KlearKall 8000 Std ROX	200 mL, contains ROX
KBS-1002-100	2X KlearKall 100 Low ROX	2.5 mL, low ROX
KBS-1002-102	2X KlearKall 1000 Low ROX	25 mL, low ROX
KBS-1001-001	2X KlearKall 100 No ROX	2.5 mL
KBS-1001-003	2X KlearKall 1000 No ROX	25 mL

Please note: The number of reactions referred to in the product name is calculated based on 50 µL total reaction volumes.

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