

OnePCR™ HiFi



Cat. No.: SM205-0100

Size: 100 Reactions (2 X 1.25 ml)

Cat. No.: SM205-0004

Size: 4 Reactions (1X 100 µl)

Description

OnePCR™ HiFi is a ready-to-use PCR reaction mixture. Simply adding primers and template, the reagent will execute primer extensions and other molecular biology applications. OnePCR™ HiFi is a pre-mixed solution containing GDP-HiFi DNA polymerase, PCR buffer, dNTPs, gel loading dyes, enhancer, and fluorescence dye. It contains the fluorescence dye, which is directly detected on the blue-light transilluminator or UV epi-illuminator after the DNA electrophoresis. The OnePCR™ HiFi mixture is supplied at the 2X concentration to allow approximately 50% of the final reaction volume to be used for the addition of the primer and template solutions. Reagents are provided with sufficient amplification reactions of 50 µl each. OnePCR™ HiFi exhibits strong proof-reading activity. The GDP-HiFi DNA polymerase exhibits excellent processivity and elongation capability. The elongation rate of this enzyme is approximately 2 times higher than that of *Taq* DNA polymerase. OnePCR™ HiFi has an extension rate of 106 to 138 nucleotides per second. OnePCR™ HiFi produces blunt end PCR products.

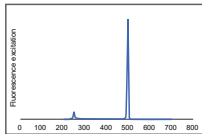


Fig. 1a. Fluorescence excitation spectra of the fluorescence dye

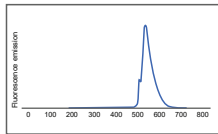


Fig. 1b. Fluorescence emission spectra of the fluorescence dye

Tracking dyes

➤ Bromophenol Blue, Xylene Cyanol FF.

Features

- No post-staining procession.
- No need to prepare PCR reagents.
- Direct loading onto your agarose gel for analysis.
- Sensitivity- High degree of sensitivity as the ethium bromide.
- Time Efficiency- No destaining requirement.
- Compatibility - Use the Blue Light or UV to detect the signal.
- Effective for the amplification of GC-rich targets.
- Exhibits strong 3'→5' exonuclease activity.

Protocol

Standard PCR with OnePCR™ HiFi:

1. For each 50 µl reaction, assemble the following components in a 0.2 ml PCR tube on ice just prior to use:

Component	Volume (µl)
OnePCR™ HiFi	25
Forward primer, 5-10 µM	Variable
Reverse primer, 5-10 µM	Variable
DNA template	Variable
Total	50

2. Mix gently. If necessary, centrifuge briefly. Cap tubes and place in the thermal cycler.
3. Process in the thermal cycler for 25~35 cycles as follows:

Initial Denaturation	2~5 minutes at 94°C	} 30 cycles
Denaturation	20~40 seconds at 94°C	
Annealing	1 minute at the proper annealing temperature	
Extension	2 mins at 72°C	
Final extension	5 mins at 72°C	

Note: Optimal conditions for amplification will vary depending on the primers and thermal cycler used. It may be necessary to optimize the system for individual primers, template, and thermal cycler.

- After the PCR reaction, DNA electrophoresis will detect the PCR product.
- Use the UV or blue-light transilluminator or UV epi-illuminator to photograph the gel.

Note: When the DNA concentration is less than 4pg, it may cause the migratory shift when performing the electrophoresis. To remedy this observation, we recommend to conduct the following steps (please refer to the experimental procedures), or use the PCR Clean-Up & Gel Extraction Kit (see NA006-0100) to remove the fluorescence dye prior to post-staining with the Novel Green (LD002-0500) or Novel Green *plus* (LD003-0500) again for restoring the DNA molecular weight in the original position.

Removal of Fluorescence Dye

- Immerse the PCR product containing the fluorescence dye into the 100 mM NaCl and add 2.5 volumes of absolute or 95% ethanol.
- Incubate on ice for 20 minutes.
- Centrifuge the mixture at 4°C for at least 10 minutes.
- Remove the suspension of ethanol and wash the pellet with 1ml of 70% ethanol.
- Dry the residual ethanol and resuspend the double-stranded DNA in the TE.

Storage

Store at RT up to 1 month.

Store at 4°C up to 6 month.

Store at -20°C up to 1 year.

Shipping Temperature: 4°C

Note: OnePCR™ HiFi is light sensitive and should be stored and protected from light.

Related Ordering Information

Cat. No.	Description	Package
BK001	BLook LED transilluminator	1 each
SM200-0100	PCR SuperMix	100 Reactions
SM201-0100	HotStar PCR SuperMix	100 Reactions
SM203-0100	OnePCR™	100 Reactions
SM206-0100	OnePCR™ HotStar	100 Reactions
SD101-0100	OneMARK 100	600 µl
SD110-0100	OneMARK B	600 µl

Caution:

- During operation, always wear a lab coat, disposable gloves, and protective equipment.
- All products are for research use only.