# amaR 2X PCR Mix



Cat. No.: SM217-0250 Size: 250 Reactions (2 × 1.25 ml)
Cat .No.: SM217-0010 Size: 10 Reactions (1 × 100 μl)

#### **Description**

The amaR 2X PCR Mix is a ready-to-use PCR reaction mixture. Simply add primers, template, and water, the reagent will execute primer extensions and other molecular biology applications. The amaR 2X PCR Mix is a pre-mixed solution containing Taq DNA polymerase, PCR reaction buffers, dNTPs, and gel loading dve. The amaR OnePCR which contains the Tag DNA polymerase, is purified from the *E. coli.*, and expressing the Thermus aquaticus DNA polymerase gene. This enzyme has a 5'  $\rightarrow$  3' DNA polymerase and the 5'  $\rightarrow$  3' exonuclease activity but lacks the 3'  $\rightarrow$  5' exonuclease activity. The amaR 2X PCR Mix can amplify DNA fragments up to 5 kb and with good amplification specificity, and it is compatible with the template. The PCR product has an A base at the 3' end and can be directly used for T/A cloning after purification. The amaR OnePCR contents red tracking dves that run at 10 bp on a 1% agarose gel. The amaR 2X PCR Mix mixture is supplied at the 2X concentration to allow 50% of the final reaction volume to be used for the addition of primer and template solutions. Reagents are provided with the sufficient amplification reactions of 20 µl each.

#### **Features**

- > No need to prepare PCR Reagents.
- > Direct loading onto your agarose gel.

# **Applications**

> PCR Amplification

#### **Kit Contents**

Contents	SM217-0250	SM217-0010
amaR 2X PCR Mix	1.25 ml X 2 vials	100 µl X 1 vial

## **Tracking Dye**

➤ Amaranth

## **Quality Control**

The quality of the amaR 2X PCR Mix is tested on a lot-to-lot basis to ensure consistent product quality.

## **Required Materials**

- > Electrophoresis equipment.
- > DNA Markers (optional).
- > DNA Staining reagent
- > BLooK LED transilluminator or UV epi-illuminator

## **Storage**

Store at room temperature up to 3 months Store at 4°C up to 6 months Store at -20°C up to 1 year Shipping temperature: 4°C

#### amaR 2X PCR Mix Protocol

#### Standard PCR with amaR 2X PCR Mix

1. For each 20 µl reaction, assemble the following components in a 0.2 ml PCR tube on ice before the

experiment.		
Component	Volume (µI)	Final Concentration
ama <i>R</i> 2X PCR Mix	10	1X
Forward primer (5-10 µM)	Variable	0.1-0.2 μM
Reverse primer (5-10 µM)	Variable	0.1-0.2 μM
DNA template	Variable	
Add ddH <sub>2</sub> O to	20	

Mix gently. If necessary, centrifuge briefly. Cap the tube and place it in the thermal cycler. 3. To process in the thermal cycler for 25-35 cycles as follows:

Initial Denaturation

Denaturation

Annealing

Extension

3-5 minutes at 94°C

30 seconds at 94°C

30 seconds at 55-65°C

30-35 cycles

30-60 seconds/kb at 72°C

Final Extension 5 minutes 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.

- After the PCR reaction, please perform DNA electrophoresis and gel staining.
- 5. Use the BLooK LED Transilluminator or UV epi-illuminator to photograph the gel.

#### **Troubleshooting**

Refer to the table below to troubleshoot problems that you may encounter when you did PCR amplification with the kit.

Problem	Cause	Solution	
Low yield of PCR products	Incomplete concentration of start materials	Use the appropriate method for the DNA preparation based on the amount of the starting materials.	
DNA degrade	DNA is not fresh	Avoid repeated freeze / thaw cycles of the sample.	
		Keep DNA preparations on ice or frozen in order to avoid the degradation.	
	DNase contaminant	Use the fresh TAE or TBE electrophoresis buffer.	
		Maintain a sterile work environment to avoid contamination from DNase.	

## **Related Ordering Information**

Cat. No.	Description	Size	
SA001-0500	AGAROSE Tablet, 0.5g	100 Tablets	
BK001	BLooK LED Transilluminator	1 Set	
SD010-R600	1 Kb DNA Ladder RTU	600 µl	
SL002-0500	Novel Green (10000X)	500 µl	
SN005-0100	Plasmid <i>mini</i> PREP Kit	100 Reactions	

#### Caution

- During operation, always wear a lab coat, disposable gloves, and protective equipment.
- > Research Use Only. Not intended for any animal or human therapeutic or diagnostic uses.