

TECHNICAL DATA SHEET:

DeCodi-Fi 2X All-in-One Mix

Product Code: DecodiFi-AIOM-2.5ml
DecodiFi-AIOM-5ml

PRODUCT DESCRIPTION

DeCodi-Fi 2X All-in-One Mix features a recombinant Hotstart High Fidelity Polymerase designed for exceptional processivity and potent proofreading capabilities. It has exceptionally low amplification bias and provides consistent sequencing coverage, delivering more reliable results.

Tailored for routine high-fidelity PCR or NGS library amplification, the DeCodi-Fi 2X All-in-One Mix empowers you with precise and robust amplification. It minimizes non-specific product formation while maximizing target yield, even when dealing with minute input quantities as low as 1 ng. Additionally, the All-in-One format provides our Hotstart High Fidelity Polymerase in a user-friendly 2X MasterMix configuration. This format includes all the necessary components for the reaction (dNTPs, MgCl₂ and stabilizers), excluding primers and template, in a proprietary buffer. Whether you're conducting routine PCR or NGS library amplification, our DeCodi-Fi 2X All-in-One Mix provides the tools you need for success.

PRODUCT APPLICATIONS

- Amplification of DNA fragments for cloning
- Amplification of long or GC rich templates
- Library amplification for sequencing

SHIPPING AND STORAGE

The DeCodi-Fi 2X All-in-One Mix has been designed by Kura Biotech to be transported within a temperature range of 2°C to 8°C without losing performance. Tests conducted have confirmed that these components can be kept under these conditions during transport for at least 7 days. Implementing these conditions in transportation helps reduce our carbon footprint by minimizing the use of thermal insulation, eliminating the need for dry ice, and simplifying logistics.

To ensure optimal kit performance, please follow the following guidelines:

- Store the components according to the specifications on each vial's label or follow the instructions in this manual. Avoid repeated freeze-thaw cycles.
- If any kit components were damaged during transportation, contact Kura Biotech. Do not use damaged or expired components as it may compromise performance.

Component	Volume 200 rxn kit	Volume 400 rxn kit	Storage T°
DeCodi-Fi 2X All-in-One Mix	2.5 µL	5 mL	-20°C

STANDARD PCR PROTOCOL

Calculate the volume of reagents needed for each reaction. Typically, reactions are set up in 25-50 μ L volumes.

General recipe for a 25 μ L reaction:

Component	25 μ L reaction	Final concentration
dH ₂ O	To 25 μ L	
DNA template	*	1ng-100 ng ^{***}
Forward primer (10 μ M)	0.5 μ L	0.2 μ M
Reverse primer (10 μ M)	0.5 μ L	0.2 μ M
DeCodi-Fi 2X All-in-One Mix	12.5 μ L	1x

Volume of template dependant on template concentration and desired input amount.

** Use \geq 5ng for gDNA, and \leq 1ng for templates \leq 50Kb

PREPARE THE PCR REACTION:

- Mix all components in a sterile PCR tube or plate and centrifuge.

PERFORM THE PCR REACTION:

- Place the PCR tubes or plates into the thermal cycler.
- Set up the cycling conditions based on the target DNA's melting temperature (T_m) and the primer characteristics.

A common PCR program consists of:

Step	Temperature	Time	Cycles
Initial denaturation	95°C	2 min	1
Denaturation	98°C	5-10 sec ^{***}	10-35*
Annealing	3-5°C below primer T_m	15-20 sec	
Extension	68°C ^{****}	30 sec ^{**} + 15 sec/kb	
Final extension	68°C	2 min	1
Hold	4°C	∞	

*Cycle numbers may need to be optimized based on specific template input, primers, and final application. Lower cycling reduces the probability of errors, and helps diminishing nonspecific products or smearing.

** Use 30 sec extension for amplicons < 1Kb, plus 10 sec/kb for longer fragments.

*** Use 98°C for 10 sec for GC-rich templates (>70% GC).

**** 68°C is preferred for avoiding depurination. 72°C can be used for amplicons <7Kb.

PCR PRODUCT ANALYSIS:

After PCR, you can analyze the products using gel electrophoresis, or you may proceed directly to downstream applications, depending on your experimental goals.