User Instruction

MAGIC MIX TRACKING DYE 2X

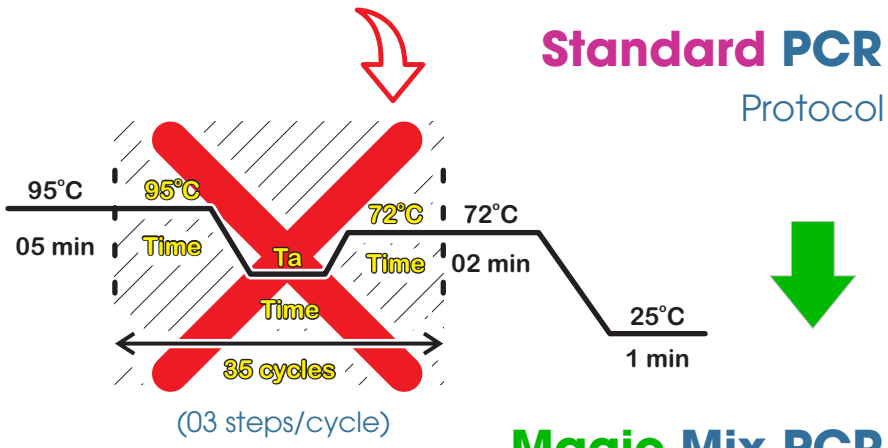
(MGM.TD 2X)

IMPORTANT: Please read carefully the user instruction before using the MGM.TD 2X

1. OVERVIEW

PHUSA Genomics MAGIC Mix MGM.TD 2X is a HOT-START enabled PCR Mix with the following specifications:

- It contains Bromophenol Blue dye to facilitate the handling of the PCR product during Agarose Gel Electrophoresis process;
- It works with 02 temperatures: 95°C for Denaturation, **Tanex (Annealing, Extension)** from 50°C to 60°C for any primers with Tm from 45°C to 70°C;
- ***The MGM.TD 2X has an optimal amplification range from 100 - 2000 bp;***
- ***The 72°C extension is no longer required.***



2. HANDLING & STORAGE:

- For shipping: It can be kept at room temperature (30°C - 37°C) for 07 days;
- For short term storage (30 days): 4°C - 10°C;
- For long term storage (01 year): -20°C - 0°C;
- For lab handling: Room temperature.

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3. PCR MIX PREPARATION

Vortex the MGM.TD 2X tube, then centrifuge gently to pull the entire solution to the bottom of the tube. Following are the recommended range of PCR reaction volumes that will allow good PCR results.

3.1 PCR reaction volume (27 - 30 uL)

Prepare the reaction mixture using the following table:

Composition	Concentration	Volume used for 1 rxn (uL)
MGM.TD 2X	2X	15
DEPC Water		10
Primer Foward	10 pmol/uL	1
Primer Reverse	10 pmol/uL	1
Template		2 - 3
Total volume		29 - 30

3.2 PCR reaction volume (20 uL)

Prepare the reaction mixture using the following table:

Composition	Concentration	Volume used for 1 rxn (uL)
MGM.TD 2X	2X	10
DEPC Water		5.5
Primer Foward	10 pmol/uL	0.75
Primer Reverse	10 pmol/uL	0.75
Template		2 - 3
Total volume		19 - 20

3.3 PCR reaction volume (15 uL)

Prepare the reaction mixture using the following table:

Composition	Concentration	Volume used for 1 rxn (uL)
MGM.TD 2X	2X	7.5
DEPC Water		3.5
Primer Foward	10 pmol/uL	0.5
Primer Reverse	10 pmol/uL	0.5
Template		2 - 3
Total volume		14 - 15

Once the PCR mix is done,

- Vortex the PCR mix, distribute into each PCR tube;
- Add 2 - 3 uL of extracted DNA to the PCR tube.
- Centrifuge gently to pull the entire solution to the bottom of the PCR tube.

4. PCR REACTION SET-UP

Tanex is the working temperature of our MGM.TD 2X, it can perform **annealing and extension** at any temperature from 40°C to 60°C.

4.1 For target up to 2000 bp

IN MULTI TARGET MODE, our MGM.TD 2X will help you to perform many PCR mix in the same run regardless of the Tm of the primers (it can vary from 45°C to 70°C).

- In **Single or Multi target** mode, if the primers Tm are ranging from 45°C-70°C, use **Tanex = 50°C** for best results;
- MGM.TD 2X allows the use of primers with 25°C differences between them.

Use the following Universal protocol for all your primers with Tm ranging from 45°C to 70°C:

Stage	Temperature	Time	Cycles
Denaturation	95°C	05 min	1
Denaturation	95°C	10 sec	30 - 35
Tanex (Annealing/ Extension)*	**50°C**	**20 sec****	
Extension	72°C	02 min	1
Keeping	25°C	30 sec	

If the primers do not produce PCR products or very low yield with smearing by-product, redo PCR with Tanex 60°C. ***Refer to Technical Guide*** *(Page 4).*

** The 72°C extension is no longer required;*

*** Tanex has been optimized for* ***20 seconds****, regardless of target length from* ***100 - 2000 bp****.*

IN MULTIPLEX MODE (several set of primers in the same PCR mix), the ***Tanex = 60°C*** should be used with your primers ***Tm varying from 50°C to 70°C***.

4.2 For target 2000 bp < length < 3000 bp

Use the following Universal protocol for all your primers with Tm ranging from 45°C to 70°C:

Stage	Temperature	Time	Cycles
Denaturation	95°C	05 min	1
Denaturation	95°C	20 sec	30 - 35
Tanex (Annealing/ Extension)*	**60°C**	**30 sec****	
Extension	72°C	03 min	1
Keeping	25°C	30 sec	

** The 72°C extension is no longer required;*

*** Tanex has been optimized for **30 seconds**, regardless of target length from **2000 - 3000 bp**.*

- THE END -

Magic Mix PCR
Technical Guide Book