DTI CobaltAmp HiFid Taq HS premix

Instruction Manual for use

User Manual for DTI CobaltAmp HiFid Taq HS premix

Catalogue number: DT0203.80, DT0203.320

Note: Applicable to all pack sizes

Manufactured by
DSS Takara Bio India Pvt Ltd
A-5 Mohan Co-operative, Industrial Estate,
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1) Product Description:

DTI CobaltAmp HiFid Taq HS premix contains a hot start PCR enzyme, optimized buffer, dNTP mixture, gel loading dye (blue), and a density reagent as a 2X premix. DTI CobaltAmp HiFid Taq HS premix is optimized for fast PCR and offers a rapid extension rate (10 sec. per kb). The inclusion of blue dye and a density reagent allows direct loading of PCR products on an agarose gel for electrophoresis. The master mix format simplifies workflows and sample handling; simply add primers, template, and water and then begin PCR.

DTI CobaltAmp HiFid Taq HS premix is ideal for fast colony PCR screening. Fast colony PCR amplification of a 5 kb insert can be completed in approximately 1 hr 15 min. Furthermore, it is possible to amplify fragments up to 6 kb from genomic DNA templates.

2) Storage :

-20℃ for long-term storage. 4℃ for short-term storage (up to 3 months).

Note: If used frequently, store at 4℃ ; the activity of the Master Mix may decrease with repeated freezing and thawing. Gently mix well before use and centrifuge briefly.

3) Applications :

- DNA amplification by PCR
- Colony PCR

4) Quality Control Data

Please see the Certificate of Analysis (CoA) for each lot.

5) Components (25 µl per reaction)

Components	Cat# DT0203.80 (80 rxn)	Cat# DT0203.320 (320 rxn)
DTI CobaltAmp HiFid Taq HS premix	1 ml	1 ml x 4
dH2O	1 ml	1 ml x 4

6) Materials required but not provided

Reagents: PCR primers, sterile purified water, template

Equipment: Thermal cycler (DTI FabSpeed thermal cycler, model# TCST-9622)

Consumables: PCR tubes, Micropipettes and tips

7) Protocol

General Reaction Composition for PCR :

Reagent	Volume	Volume
DTI CobaltAmp HiFid Taq HS premix	12.5 µl	25 µl
Forward Primer	0.1 µM (final conc.)	0.2 µM (final conc.)
Reverse Primer	0.1 µM (final conc.)	0.2 µM (final conc.)
Template - human genomic DNA	50 - 100 ng	50 - 100 ng

- plasmid DNA	100 pg - 10 ng	100 pg - 10 ng
dH2O	up to 25 µl	up to 50 µl
Total	25 µl	50 µl

8) Recommended PCR Conditions :

3 step PCR (human or mouse genomic DNA : up to 2 kb product, bacterial genomic DNA or colony PCR insert : up to 6 kb product)

94℃, 1 min →	98℃	5 sec	⎫
	55℃*	5 sec	⎬ 30 cycles
	72℃	10 sec/kb*	⎭

* 5 sec for less than 1 kb

2 step PCR (human or mouse genomic DNA : 2 - 6 kb)

94℃, 1 min →	98℃	5 sec	⎫ 30 cycles
	68℃	30 sec/kb	⎭

NOTE: Denaturation conditions vary depending on the thermal cycler and tubes used for PCR. We recommend denaturing for 5 - 10 sec at 98℃ or 20 - 30 sec at 94℃.

9) PCR product :

Since most PCR products amplified with DTI CobaltAmp HiFid Taq HS premix have an A overhang added at 3'-termini, the obtained PCR product can be used directly for cloning into a T-vector. Additionally, it is possible to clone the product in a blunt-end vector after blunting and phosphorylation of the end.

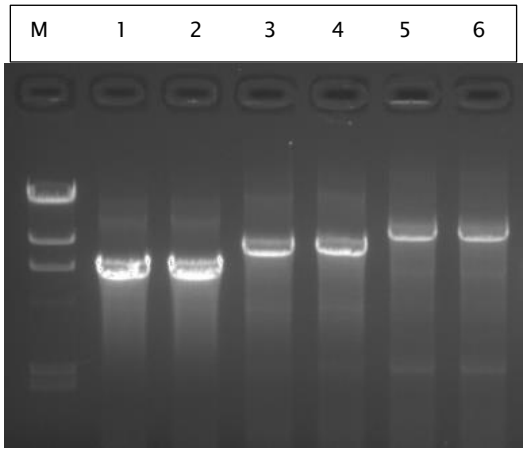
10) Dye marker migration during Electrophoresis :

When 5 µl of the PCR sample is loaded on a 1% gel made with DTI agarose routine (cat# DT1701.500) and subjected to electrophoresis, the blue dye fronts are detected at positions corresponding to 1 kb and 3 - 5 kb. The absorption maxima for the dyes are ~260 nm and 620 nm, respectively. The dyes may be removed by isolating and purifying the DNA fragment from the gel or extracting DNA with NucleoSpin Gel and PCR Clean-up (Cat. #740609.50/.250), if necessary.

11) Experimental sample:

Consistent amplification of 6 kb, 8 kb, 10 kb fragments from E.coli genomic DNA is observed using the DTI CobaltAmp HiFid Taq HS premix. The protocol used for the assay and the results are as follows:

94 C	1 min	
98 C	5 sec	30 Cycles
55 C	5 sec	
72 C	1 min 40 sec	



Lane M: λ Hind III digest marker
Lane 1-2: Test in duplicates- 6 kb
Lane 3-4: Test in duplicates- 8 kb
Lane 5-6: Test in duplicates- 10 kb

Figure 1: Gel electrophoresis image of PCR

Visit https://store.dsstakarabio.com/ for more detailed product information

For more information contact directly below;
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Description of Symbol Used:

REF Catalogue number

LOT Batch Code

Date of Manufacturing

Use-by-date

Contains sufficient for <n> tests