IsoFast® **Hot Start Bst Colour Mix**

- Sensitive & specific
- AptaLock™ hot start
- Fast colourimetric readout

IsoFast® Hot Start Bst Colour reagents are colourimetric isothermal amplification enzyme formulations that combine IsoFast® Hot Start Bst Polymerase with a pH-based dye for rapid positive/negative screening.

Features

- Fast colour readout for positive/negative testing
- AptaLock™ hot start for ultra-sensitive detection of DNA targets
- Rapid polymerisation for faster time to result (as little as 10 mins)
- Detect down to 3 target copies per µL
- Ideal for both cold and room temperature setup
- Improved speed and sensitivity for early target detection
- High activity at a broad range of temperatures from 55-70 ˚C

Applications

- Whole genome amplification
- Multiple displacement amplification
- Isothermal amplification
- LAMP-type amplification
- Molecular diagnostic test developement
- Point-of-care testing

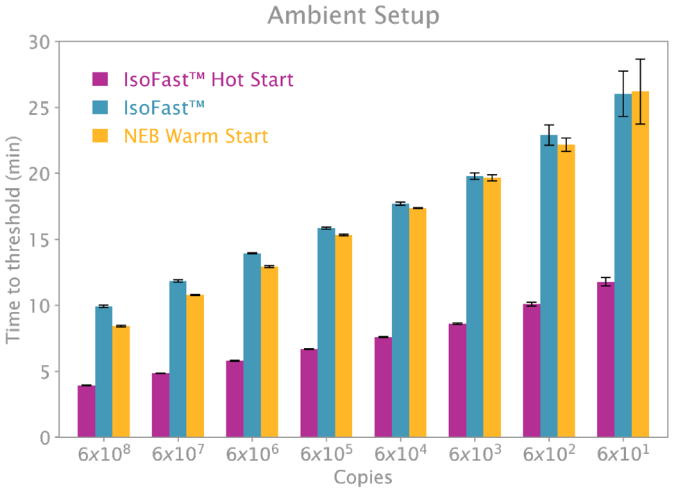

Figure 1. Improved time to result under ambient setup

Isothermal amplification of a target sequence in the scaffolding protein gene of the M13 bacteriophage genome using IsoFast® Hot Start Bst Mix, IsoFast® Bst Mix, and NEB WarmStart LAMP Kit. A primer mix consisting of 0.2 µM for F3 and B3 primers, 1.6 µM for FIP and BIP primers and 0.8 µM for LoopF and LoopB primers was used. The total reaction volume was 25 µL. 8 serial dilutions of M13 ssDNA genome were used, starting with a stock of 0.5 ng/µL and using a dilution factor of 10, corresponding to the number of genome copies indicated in the plot. Reaction master mixes and plates were prepared at room temperature (ambient setup), for approximately 20 min. The reaction was run at 65 ˚C for 100 minutes. A BioRad CFX96 Touch instrument was used to record fluorescence every 10 seconds. The time to threshold indicates the time required to reach the same fluorescent threshold.

IsoFast® Hot Start Bst Mix shows faster amplification when compared to IsoFast® Bst Mix and to NEB WarmStart LAMP Kit, under ambient setup.

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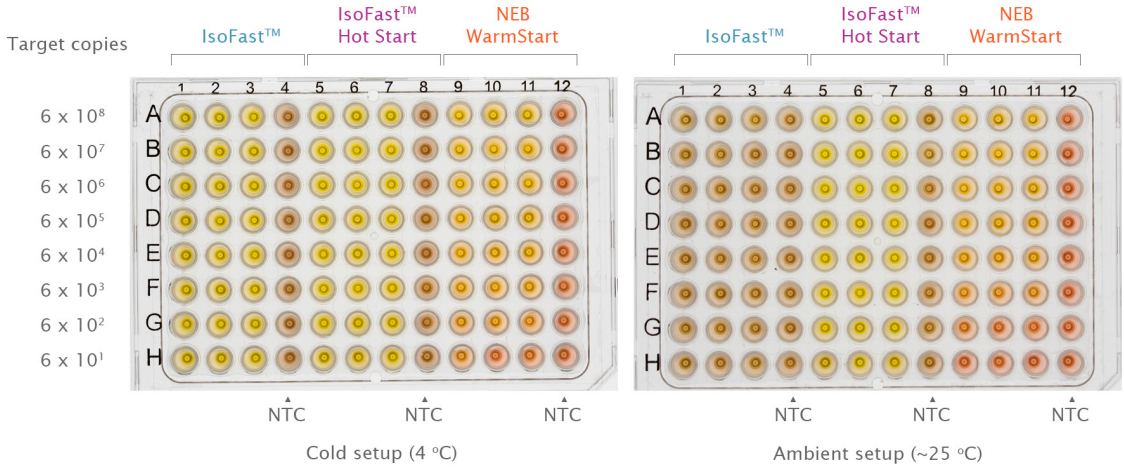

Figure 2. Reliable colourimetric readout with cold and ambient reaction setup

Isothermal amplification of a target sequence in the scaffolding protein gene from the M13 bacteriophage genome using IsoFast® Hot Start Bst Polymerase or IsoFast® Bst Polymerase in 10x IsoFast® Colour Buffer A, and NEB WarmStart Colorimetric LAMP 2X Master Mix. A primer mix consisting of 0.2 µM for F3 and B3 primers, 1.6 µM for FIP and BIP primers and 0.8 µM for LoopF and LoopB primers was used. The total reaction volume was 25 µL. 8 serial dilutions of M13 ssDNA genome were used, starting with a stock of 0.5 ng/µL and using a dilution factor of 10, corresponding to the number of genome copies indicated next to the plates. Reaction master mixes and plates were prepared either using cold blocks (cold Setup) or at room temperature (ambient setup), for approximately 20 min. The reaction was run at 65 ˚C for 30 minutes. Plates were then photographed to show the colours obtained at the end of the run. IsoFast® Bst Polymerase and IsoFast® Hot Start Bst Polymerase showed a better sensitivity compared to NEB WarmStart Colorimetric LAMP 2X Master Mix in Cold Setup.

IsoFast® Hot Start Bst Polymerase allows easy screening for positives even with ambient temperature setup.

Hot start isothermal amplification

IsoFast® Hot Start Bst Polymerase is a recombinant version of the large fragment of *Geobacillus stearothermophilus* (formerly known as *Bacillus stearothermophilus*, Bst) DNA polymerase. It is engineered to retain 5'-3' polymerase activity, but lacks 5'-3' exonuclease activity.

The enzymes' strong strand displacement capability eliminates the need for denaturation required by standard PCR. Combined with powerful hot start technology, to minimise primer dimer formation and non-specific target amplification, this novel mix is ideal for colourimetric LAMP assays.

Reliable positive/negative testing

IsoFast® Hot Start Bst Colour Mix outperforms the competitor mix at both cold and ambient setup. The mix enables detection of down to 3 copies per microlitre, with a clear colour change, compared to the no-template control. Unlike the competitor mix, no reactions resulted in false negatives. High test speeds and accurate results make this the reagent of choice for point-of-care testing and field-based screening.

Catalogue Number	Product Name	Pack Size	Presentation
PB80.50-01	IsoFast® Hot Start Bst Polymerase Colour	1600 Units	[1 x 200 µL 8 U/µL] & [1 x 500 µL Colour Buffer A] & [1 x 1 mL Buffer B]
PB80.50-08		8000 Units	[1 x 1 mL 8 U/µL] & [2 x 1.25 mL Colour Buffer A] & [3 x 1.7 mL Buffer B]
PB80.51-01	IsoFast® Hot Start Bst Colour Mix	100 Reactions	1 x 1.25 mL
PB80.51-05		500 Reactions	5 x 1.25 mL

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