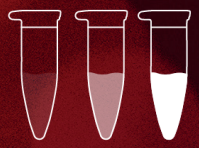
IsoFast® **Bst 1-Step Mix**



- Fast
- High yield
- Versatile

IsoFast® Bst 1-Step Mix is a dual enzyme system for rapid and sensitive isothermal amplification of RNA targets in one step. The kit contains IsoFast® Bst Polymerase, which provides strong strand displacement capabilities, together with the highly active modified MMLV RTase Go.

Features

- Validated for qualitative detection of SARS-CoV-2 nucleic acid
- Includes strand-displacing IsoFast® Bst Polymerase in a 2x mix format
- Supplied with RTase Go plus RNase inhibitor
- Reaction is carried out at a constant temperature (65 ˚C)
- Gives rapid and consistent amplification across a wide range of templates
- Includes an advanced buffer system for higher yield under difficult conditions
- Supplied with a fluorescent dye for real-time detection
- 30 minute protocol

Applications

- Multiple displacement amplification
- Isothermal amplification
- Loop mediated isothermal amplification (LAMP)
- Molecular diagnostics
- Field diagnostics

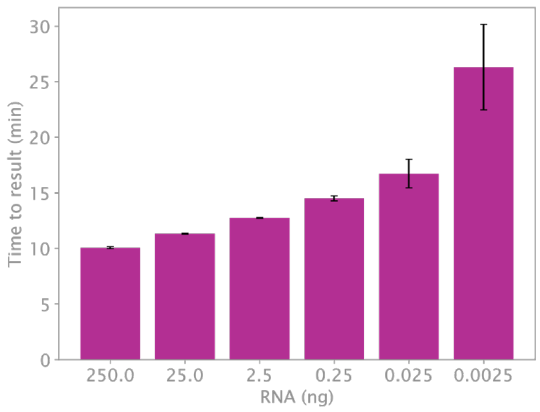


Figure 1. Rapid and sensitive amplification performance

Isothermal amplification of beta actin from human lung total RNA using IsoFast® Bst 1-Step Mix. A primer mix 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. 7 serial dilutions of template were used, corresponding to 250 ng, 25 ng, 2.5 ng, 250 pg, 25 pg, 2.5 pg and 250 fg of total RNA. The reaction was run at 65 ˚C for 34 minutes. A BioRad CFX96 Touch instrument was used to record fluorescence every 10 seconds. Time to result is the time required to reach the same fluorescent threshold. IsoFast® Bst 1-Step Mix provides rapid and sensitive amplification down to 2.5 pg of total RNA.

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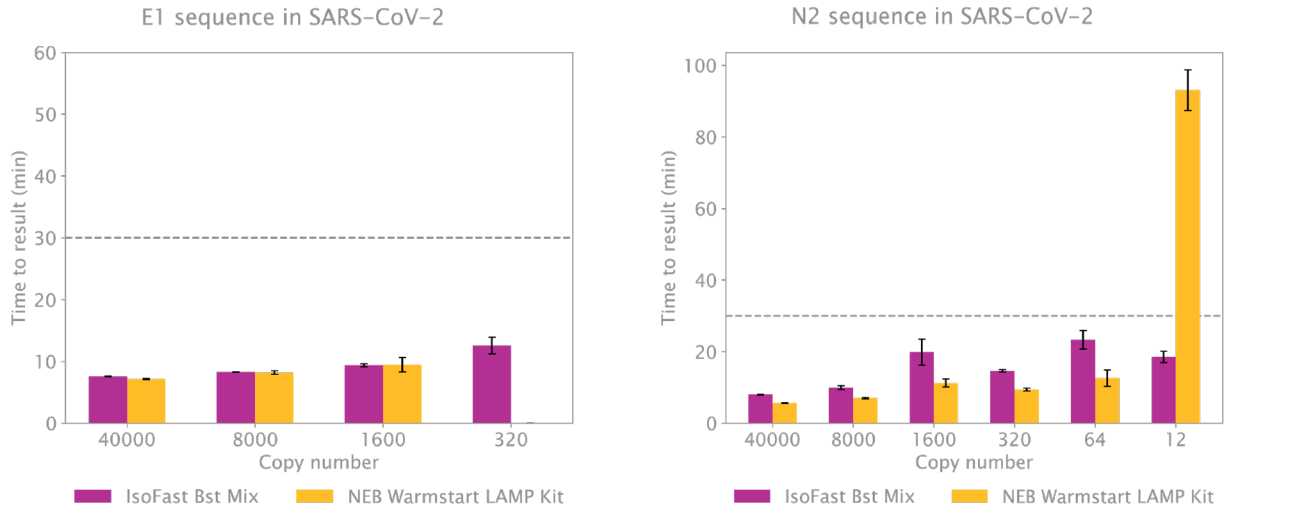

Figure 2. Rapid detection of SARS-CoV-2 E1 and N2 sequence

Isothermal amplification of E1 and N2 targets in SARS-CoV-2 RNA using IsoFast® Bst 1-Step Mix and compared to results obtained with NEB WarmStart® LAMP Kit. A primer mix of 0.2 µM for F3 and B3, 1.6 µM for FIP and BIP and 0.8 µM for LoopF and LoopB primers was used. The total reaction volume was 25 µL. 7 serial dilutions of template were used, corresponding to 40,000, 8000, 1600, 320, 64, 12.8, 2.56 copies of SARS-CoV-2 RNA. The reaction was run at 65 ˚C for 100 minutes. A BioRad CFX96 Touch instrument was used to record fluorescence every 10 seconds. Time to result is the time required to reach the same fluorescent threshold. IsoFast® Bst 1-Step Mix enables rapid detection of all dilutions of SARS-CoV-2 RNA tested.

Dual enzyme system

IsoFast® Bst 1-Step Mix is designed for reverse transcription of target RNA and subsequent isothermal amplification in a single tube.

The kit utilises IsoFast® Bst Polymerase for its strong strand displacement activity, enabling DNA synthesis at a constant temperature (65˚C) without the need for thermal cycling. Representing the large fragment of *Geobacillus stearothermophilus* (formerly *Bacillus stearothermophilus*) DNA Polymerase, this portion of the protein catalyses the 5'-3' synthesis of DNA but does not contain the 5'-3' exonuclease domain.

Reverse transcription of target RNA is carried out by the thermostable and extremely active RTase Go, which is blended with RNase inhibitor to prevent degradation of RNA by contaminating RNase.

Rapid and consistent results

Designed for fast amplification, IsoFast® Bst 1-Step Mix gives rapid and consistent results across a wide range of sample types and is validated for qualitative detection of SARS-CoV-2 nucleic acid (Fig 2). The buffer chemistry ensures high yield and performance even under difficult conditions, for example when inhibitors are present.

IsoFast® Bst 1-Step Mix requires only the addition of primers, template and water. The kit is provided with a separate tube of fluorescent dye to allow real-time detection with any qPCR instrument.

Cat. No.	Product Name	Pack Size	Presentation
PB80.21-01	IsoFast® Bst 1-Step Mix	100 Reactions	[1 x 1.25 mL Bst Mix] & [1 x 200 µL RTase Go] & [1 x 125 µL Dye]
PB80.21-05		500 Reactions	[4 x 1.56 mL Bst Mix] & [1 x 1mL RTase Go] & [1 x 625 µL Dye]

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