A new generation of hot-start polymerase that delivers improved specificity, yield, speed and robustness when amplifying targets from any DNA template.

MyTaq™ HS is a new generation of antibody-mediated hot-start enzyme, engineered for highly specific and efficient amplification from even the most challenging templates. MyTaq HS remains inactive at room temperature allowing for convenient reaction set-up, thereby reducing non-specific amplification that can hinder PCR assays from the start. These properties make MyTaq HS DNA Polymerase the ideal choice for PCR assays containing complex and low copy number targets, as well as multiplex PCR.

MyTaq™ HS Mix is comprised of MyTaq HS DNA Polymerase and a novel buffer system, that requires only the addition of template, primers and water, thereby reducing the number of pipetting steps during PCR set-up for improved speed, throughput and assay reproducibility.

The inclusion of dNTPs, MgCl₂ and enhancers at optimal concentrations, helps eliminate the need for optimization, thereby saving time, effort and the cost of performing unnecessary assay repeats.

APPLICATIONS
MyTaq HS has been validated with a full range of templates and is perfectly suited to the following applications:

- Fast PCR
- Multiplex PCR
- Genotyping
- Complex templates (e.g. GC-rich)
- Colony PCR
- Low copy number PCR assays
- High-throughput assays with prolonged set-up

HIGHER YIELDS
MyTaq HS DNA Polymerase has been developed to give more robust amplification than other commonly-used polymerases, meaning it performs reliably even in the presence of PCR inhibitors (Fig. 1), permitting amplification of longer fragments (Fig.2). An example is in colony PCR, where conventional polymerases are easily inhibited by bacterial cell debris and components of the cell culture media, often resulting in bias, where the amplification of larger amplicons is hindered.
FAST, HIGH-THROUGHPUT PCR

MyTaq HS only requires one minute activation and allows fast cycling conditions to be used, reducing the reaction time to under 30 minutes, without compromising PCR specificity or yield (Fig. 3). This makes MyTaq HS suitable for both routine and high-throughput PCR.

The speed of MyTaq HS was further reduced using an ultra-fast protocol, in which MyTaq HS amplified a 900 bp fragment of genomic DNA in under 20 minutes (Fig. 4).

Fig. 1 Robustness of MyTaq HS in colony PCR
A 2.6 kb fragment of human genomic DNA was cloned into M13 and transformed into E. coli cells. 1 mL of a 1:16 dilution of an overnight culture of these cells was used directly in a 50 µL PCR reaction.

A) 2 µL increments of agar were added (Lanes 1 - 8 respectively, HyperLadder 1kb (M)). B) 2 µL increments of LB were added (Lanes 1 - 8 respectively, HyperLadder 1kb (M)) and amplified according to manufacturers' protocol. MyTaq HS DNA Polymerase was more resistant to inhibition than the polymerase from Supplier S, making it ideal for colony PCR and liquid overnight cultures, offering improved workflows particularly for high throughput assays.

Fig. 2 Colony PCR of long fragments
E. coli transformed with M13 carrying the 2.6 kb or an 884 bp insert was plated out. 12 colonies were picked with tooth-picks, washed directly into MyTaq buffer and amplified using MyTaq HS. HyperLadder 1kb (M). Fragments up to 3 kb can be reliably amplified using fast cycling conditions with MyTaq HS DNA Polymerase. This illustrates the ability of MyTaq HS to amplify full-length cloned inserts and facilitates the rapid identification of correct plasmid inserts.

Fig. 3 Fast amplification was carried out on a range of human genomic targets
A 340 bp (A) and a 450 bp (B) fragment of the myc gene, a 525 bp (C) fragment of the EGFR gene and a 530 bp (D) fragment of the AGTR1 gene was amplified using MyTaq HS polymerase and the results were compared with PCR reactions using hot-start DNA polymerases from other suppliers. A 3-fold serial dilution of human genomic DNA (100 ng - 3 pg, lanes 1 - 8 respectively (HyperLadder 1kb (M)) was incubated for 3 min at 95 °C followed by 35 cycles of 95 °C for 15s, 55 °C for 15s and 72 °C for 15s . MyTaq has performed well across all four human genes.
PREMIXES FOR INCREASED REPRODUCIBILITY

MyTaq HS Mix and MyTaq HS Red Mix contain all the required components for easy PCR set-up. Both MyTaq HS Mix and MyTaq HS Red Mix are conveniently supplied in one tube, maximizing convenience, facilitating greater.

MULTIPLEX PCR

The key to successful multiplex PCR is the ability to define a set of common reaction parameters to all of the primer sets in the reaction, to ensure highly-specific target annealing efficiency and reproducibility. MyTaq HS Mix is based on the latest technology in PCR enzyme preparation, the pre-configured reaction mix contains a unique combination of salts and additives to ensure comparable efficiencies for annealing and extension of all primers in the reaction. This makes MyTaq HS Mix ideal for multiplex PCR (Fig. 5).

DIRECT GEL LOADING

MyTaq HS is also supplied as MyTaq HS Red DNA Polymerase and MyTaq HS Red Mix, which includes a 5x MyTaq Red Reaction Buffer that increases the visual contrast between the reagent and the reaction vessel for improved convenience and to improve pipetting accuracy. The red dye also enables samples to be loaded directly on to a gel after the PCR eliminating the need to add loading buffer.

Fig. 4 Ultra-fast amplification of the human AGTR1 gene

A 900 bp fragment of the AGTR1 gene was amplified with MyTaq HS Mix and hot-start Taq polymerases from other suppliers. A 3-fold serial dilution of human genomic DNA (100 ng - 3 pg, lanes 1-8 respectively) was used and incubated at 95 °C for 3 min, followed by 35 cycles of 95 °C for 5 s, 55 °C for 1s and 72 °C for 15 s. Only MyTaq HS was capable of amplifying a 900 bp fragment of human genomic DNA under such fast conditions.

Fig. 5 Successful 16-plexing using MyTaq HS Mix

50 ng of human genomic DNA was used as a template in 25 µL PCR reactions, with primers to amplify 14 individual amplicons: 335 bp, 355 bp up to 961 bp (lanes 1 - 16 respectively) and B) 1-, 2-, 3-, 6-, 8-, 10-, 12-, 14-, and 16-plex reaction (lanes 1-9 respectively). The cycling was performed under the recommended multiplex conditions: 95 °C for 2 min, followed by 25 cycles of 95 °C for 30 s, 65 °C for 4 min. These results demonstrate that MyTaq HS Mix can be used successfully for multiplex PCR without the need for extensive optimization.
Certain PCR can be problematic if the Taq is not top quality. We had barely any signal with other Taq. When we switched to MyTaq HS, the bands were there and clean! Nice product!

University of California, San Francisco (UCSF), US

Ordering Information

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For related products such as nucleotides, agarose and molecular weight markers visit www.bioline.com