Nova HotTaq Premix

2X concentrated

Reference: AB12039; AB12040; AB12041

INTENDED USE AND PRESENTATION:

This HotTaq Premix is a hot start version widely used in molecular biology.

AB12039, 1 mL. 1 mL.

AB12040, 3 mL. 1 mL x 3.

AB12041, 5 mL. 1 mL x 5.

One unit is defined as the amount of enzyme required to catalyze the incorporation of 10 ηmoles of dNTPs into acid-insoluble material in 30 minutes at 74°C.

For research use only.

SUMMARY, EXPLANATION AND LIMITATIONS:

Nova HotTaq 2x PCR Premix is a premixed solution containing everything needed for successful PCR reaction except specific primers and DNA template. The premix includes high-quality recombinant Nova HotTaq DNA Polymerase, nucleotides and magnesium in a PCR reaction buffer.

For the reaction set-up add the PCR Premix (10 or 25 µl) to the primers, template and water for the total reaction volume of 20 or 50 µl. To activate the Nova HotTaq DNA Polymerase it should be incubated at 95 - 97o C for 15 minutes as a first PCR step.

This enzyme allows the PCR setup at ambient temperature without nonspecific annealing and extension.

Quality control: Activity and stability tested at 20, 30 and 40 cycles of PCR reactions at 95°C. Free of detectable, non-specific nucleases.

Several cycles of freezing/thawing are allowed.

APPLICATIONS:

Hot-start PCR, RT-PCR, Amplification of low copy or high range size DNA targets, Real-Time PCR, Multiplex PCR, T-vector cloning.

PRODUCT COMPOSITION:

-Nova HotTaq DNA Polymerase 0,1 unit/µL in reaction buffer, 4 mM $MgCl_2$, 0,4 mM of each dNTPs (dATP, dCTP, dGTP, dTTP).

- Free nuclease water.

METHODS AND PROCEDURE:

Optimal reaction conditions, such as reaction time, temperature, and amount of template DNA, may vary and must be individually determined.

General Reaction Protocol:

1. Thaw 2X Nova HotTaq Premix solution.

2. Prepare a master mix.

Recommended PCR reaction mix:

Component	Volume	Final Conc.
2X Nova HotTaq Premix	10 µL	1X
Upstream Primer (10 pmoles/µL)	Variable	0,3-1,0 µM
Downstream Primer (10 pmoles/µL)	Variable	0,3-1,0 µM
Template DNA	Variable	1-100 ηg/µl
Sterilized D.W.	Up to 20 µL	-
Total Volume	20 µL	-

*Primers should have a predicted melting temperature of around 60°C, using default Primer 3 settings (http://frodo.wi.mit.edu/primer3/).

Amount of template:
Bacteriophage λ, cosmid, plasmid → 10 fg~300 ηg
Total genomic DNA → 100 ηg~1 µg

3. Mix the master mix thoroughly and dispense appropriate volumes into PCR tubes. Centrifuge the reactions in a microcentrifuge for 10 seconds.

4. Perform PCR using your standard parameters (3-step cycling).

Step	Temperature & Reaction Time		
Initial denaturation	15 min. at 95°C	-	-
DNA amplification 25-35 cycles	30-60 sec. at 95°C	30-60 sec. at 50-68°C	1-4 min. at 72°C
Final extension	-	-	5-10 min at 72°C

*Annealing temperature should be 2-6ºC lower than the primer melting temperature.

5. Separate the PCR products by agarose gel electrophoresis and visualize with EtBr or any other means.

A DNA fragment which is amplified by HS Prime Taq DNA Premix has A overhang, and it enables you to do cloning by using T-vector.

REQUIRED MATERIALS BUT NOT SUPPLIED:

All reagents, materials, and laboratory equipment for qPCR procedures are not provided with this polymerase. This includes sterile reaction tubes, micropipettes and tips, template DNA, gen-specific PCR primer pair, dNTPs mixture, PCR grade H_2O, heat pretreatment equipment (thermoblock, microwave), centrifuge, cold store and thermal block cycler.

Buffered solutions for DNA extraction or purification, enzyme treatments, highly sensitive detection systems, and other auxiliary reagents are available from Gennova Scientific.

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STORAGE AND STABILITY:

Store at -20°C until the expiration date printed on product label. Avoid prolonged exposure to light. Avoid multiple freeze-thaw cycles and exposure to frequent temperature changes. Do not use after the expiration date. If the product is stored under different conditions from those stipulated in these technical indications, the new conditions must be verified by the user. The validity period of the ready to use products when opened, is the same as the expiration date indicated on the label of intact product.
Gennova Scientific guarantees that the product will maintain all of the described characteristics from the production date until the expiration date, as long as the product is stored and used as recommended. No other guarantees are provided. Under no circumstances Gennova Scientific is obliged to cover damages caused by use of this reagent.

TROUBLESHOOTING:

If unusual amplification is observed or any other deviations from the expected results, please read these instructions carefully, along with the instructions from the PCR system. If this does not solve the problem, please contact Gennova Scientific's technical support department (techsupport@gennovalab.com) or your local distributor.

PRECAUTIONS:

Use only by qualified personnel.
Use proper protective equipment in order to avoid contact with reagents and samples in the eyes, skin, and mucosal tissues. In case of contact with sensitive areas, immediately flush the affected area with water. Avoid microbial contamination of the reagent, as this may produce nonspecific amplification results.
Material safety data sheet (MSDS) is available upon request.

PERFORMANCE CHARACTERISTICS:

Gennova Scientific has performed studies to evaluate the functioning of this polymerase for use with standard amplification systems, concluding that the product is both specific and sensitive for PCR performance.

BIBLIOGRAPHY:

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Lo Y.M., Mehal W.Z., Fleming K.A., "Rapid production of vector-free biotinylated probes using the polymerase chain reaction", Nucleic Acids Research, 16(17), 8719, 1988.
Erlich H.A., (ed.) 1988, "PCR technology: principles and applications for DNA amplification", Stockton Press, New York.

REF Catalog number
LOT Batch code
RUO Research use only
Temperature limitation
Expiration date
Manufacturer
See instruction for use

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