

EcoNova HotTaq Premix Ready-to-Load

5X concentrated, 12,5 mM

Reference: AB12030; AB12031; AB12032



1 of 2

INTENDED USE AND PRESENTATION:

This HotTaq Premix is widely used in molecular biology research.

AB12030, 1 mL. 1 mL.

AB12031, 3 mL. 1 mL x 3.

AB12032, 5 mL. 1 mL x 5.

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

For research use only.

SUMMARY, EXPLANATION AND LIMITATIONS:

EcoNova HotTaq PCR Mix Ready-to-Load is a premixed ready-to-use solution containing: all reagents required for PCR (except template, primers and water), additional compound needed for direct loading onto agarose gel and two tracking dyes (blue and yellow) that allow to monitor progress during electrophoresis.

EcoNova HotTaq Premix contains two enzymes – EcoNova HotTaq DNA polymerase and a proofreading polymerase.

These enzymes together have both the 5' \rightarrow 3' exonuclease activity as well as the 3' \rightarrow 5' proofreading activity.

Reaction setup at room temperature is highly recommended for EcoNova HotTaq Premix.

We recommend using EcoNova HotTaq Premix in any PCR application that will be visualized by agarose gel electrophoresis and NovaSight DNA Stain or Ethidium bromide staining.

Quality Control: Both Endonuclease and Exonuclease activities have been detected.

APPLICATIONS:

EcoNova HotTaq Premix Ready-to-Load is a premix for all your everyday PCR reactions, gene expression profiling, Microbial & Viral pathogen detection. This EcoNova Premix allows efficient amplification from GC-rich and AT-rich templates, under fast and standard cycling conditions.

PRODUCT COMPOSITION:

-EcoNova HotTaq DNA Polymerase

-Proofreading enzyme

-5X reaction buffer

-12,5 mM MgCl₂

-2 mM dNTPs of each

-BSA

-Blue dye: Migration equivalent to 3,5-4,5 kb DNA fragment

-Yellow dye: Migration rate in excess of primers in 1% agarose gel: <35-45 bp

-Compound that increases sample density for direct loading

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 5X EcoNova HotTaq Premix Ready-to-Load.

2. Prepare a master mix.

Recommended PCR reaction mix:

Component	Volume	Final Conc.
5X EcoNova HotTaq Premix RTL	4 μ L	1X
Upstream Primer (10 pmoles/ μ L)	0,2-0,6 μ L	0,1-0,3 μ M
Downstream Primer (10 pmoles/ μ L)	0,2-0,6 μ L	0,1-0,3 μ M
Template DNA	Variable	5-50 ng/ μ L
Sterilized D.W.	Up to 20 μ L	-
Total Volume	20 μ L	-

*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 \rightarrow 10 fg~300 ng

Total genomic DNA \rightarrow 100 ng~1 μ g

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	12-15 min. at 95°C	-	-
25-35 cycles	10-20 sec. at 95°C	30-60 sec. at 54-66°C	20 sec.-4 min. at 72°C
Final extension	-	-	5-10 min at 72°C

***IMPORTANT:** To activate the polymerase, include an incubation step at 95°C for 12 - 15 minutes at the beginning of the PCR cycle.

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

A DNA fragment which is amplified by EcoNova Taq DNA Polymerase 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 H₂O, 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.



Catalog number



Batch code



Research use only



Temperature limitation



Expiration date



Manufacturer



See instruction for use



Gennova Scientific, S.L.
C/ Johann Gutenberg, 4F. Pol. Ind.
El Cafamo I • 41300 San Jose
de La Rinconada • Sevilla, SPAIN
Telefono: +34 954 150767
Fax: +34 955 266494

info@gennovalab.com
www.gennova-europe.com

EcoNova HotTaq Premix Ready-to-Load

5X concentrated, 12,5 mM

Reference: AB12030; AB12031; AB12032



2 of 2

STORAGE AND STABILITY:

Shipping and temporary storage for up to 1 month at room temperature or storage for up to 6 months at 2-8°C has no detrimental effects on the quality of EcoNova HotTaq Premix. 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:

Chien A., Edgar D.B., Trela J.M., "Deoxyribonucleic acid polymerase from the extreme thermophile *Thermus aquaticus*", *Journal of Bacteriology*, 127(3), 1550-57, 1976.
Lawyer F.C., Stoffel S., Saiki R.K., Myambo K., Drummond R., et al., "Isolation, characterization, and expression in *Escherichia coli* of the DNA polymerase gene from *Thermus aquaticus*", *The Journal of Biological Chemistry*, 264(11), 6427-37, 1989.
Tindall K.R., Kunkel T.A., "Fidelity of DNA synthesis by the *Thermus aquaticus* DNA polymerase", *Biochemistry*, 27(16), 6008-13, 1988.
Innis M.A., Myambo K.B., Gelfand D.H., Brow M.A., "DNA sequencing with *Thermus aquaticus* DNA polymerase and direct sequencing of polymerase chain reaction-amplified DNA", *Proceedings of the National Academy of Sciences of the United States of America*, 85(24), 9436-40, 1988.
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.
Erich H.A., (ed.) 1988, "PCR technology: principles and applications for DNA amplification", Stockton Press, New York.

F01T04_AB12030_AB12031_AB12032_V1R1012_EN_EcoNova_HotTaq_Premix_RTL



Catalog number



Batch code



Research use only



Temperature limitation



Expiration date



Manufacturer



See instruction for use



Gennova Scientific, S.L.
C/ Johann Gutenberg, 4F. Pol. Ind.
El Cádizamo I • 41300 San José
de La Rinconada • Sevilla, SPAIN
Teléfono: +34 954 150767
Fax: +34 955 266494

info@gennovalab.com
www.gennova-europe.com