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HGS Diamond *Taq*® DNA Polymerase

Specification Sheet

Reference: TAQ-I011

Eurogentec products are sold for research or laboratory use only and are not to be administrated to humans or used for medical diagnostics. For medical diagnostics, please use the TAQ-I010 references.

Source

HGS Diamond *Taq*® is a highly thermostable enzyme produced and purified from recombinant *Escherichia coli* bacterium containing the *Thermus aquaticus* DNA Polymerase gene.

Intended use

HGS Diamond *Taq*® lacks *Taq* I restriction endonuclease activity. The enzyme shows very good fidelity and catalyzes 5'→3' polymerization-dependent exonuclease replacement activity with no detectable 3'→5' exonuclease activity. HGS Diamond *Taq*® is a chemically modified Hot Start *Taq* DNA polymerase, which completely lacks any activity before activation to avoid non-specific priming at low temperature. This enzyme requires a 10 minutes activation step at 95 °C to reach maximal initial activity. During the PCR the rest of its activity is released. It is heat-degraded at a much lower rate as commonly used *Taq* DNA polymerase. DNA fragments as long as 2 kb can be efficiently amplified. HGS Diamond *Taq*® DNA polymerase provides efficient amplification of specific products without amplifying non-specific products or primer dimers. HGS Diamond *Taq*® is particularly suited for diagnostic PCR & qPCR applications that require high sensitivity and ultra low levels of bacterial & fungal and/or highly specific amplification. The GMP manufacturing & purification processes minimize the risk of false positive results due to residual DNA contamination (bacterial or fungal). The enzyme is QC-tested to verify that < 1fg of genomic *E. coli* DNA (or 0.2 copy) is present in a standard aliquot containing 1 unit of Taq. Bioburden is guaranteed ≤ 10 CFU/ml, but is typically = 0 CFU/ml.

Package contents

Reference	Units	Volume	Concentration	Volume HGS Diamond Taq® reaction buffer (10 X)*	Volume 25 mM $MgCl_2$
TAQ-I011-100 (sample)	100	20 µl	5 U/µl	1 ml	1 ml
TAQ-I011-1000	1000	200 µl	5 U/µl	6 ml	6 ml
TAQ-I011-5000	5000	1 ml	5 U/µl	30 ml	30 ml
TAQ-I011-25000	5 x 5000	5 x 1 ml	5 U/µl	5 x 30 ml	5 x 30 ml

*150 mM Tris-HCl pH 8.5 (at 19 °C), 500 mM KCl and stabilizers.

Shipping conditions

Shipping at room temperature

Storage conditions

Storage at -20°C is recommended

Storage and dilution buffer

20 mM Tris-HCl, 1 mM DTT, 0.1 mM EDTA, 0.1 M KCl, 0.5% (v/v) Nonidet P40, 0.5% (v/v) Tween 20, 50% (v/v) glycerol, pH 8.0 (19°C) and stabilizer.

Enzyme Specifications

Each lot of enzyme, buffer and $MgCl_2$ is functionnaly tested and quality controlled to ensure the following specifications of the IVD-GMP products.

Appearance	Colourless solution
Identity	MW approx. 95 kDa
Volume activity	≥ 5 Units/µl
Specific activity	NA
Purity	> 98%
Performance test on λ DNA (PCR)	0.5 kb fragment positive down to 5 pg
Performance test on 18S DNA (PCR)	0.1 kb fragment positive down to 10 pg
Absence of Ribonucleases	Not detectable up to 10 U, 1 h, 37°C
Absence of Endonucleases	Not detectable up to 30 U, 16h, 65°C
Absence of Exonucleases	Not detectable up to 30 U, 16h, 65°C

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Absence of Nicking activity	Not detectable up to 30 U, 16h, 65°C
5'-3' exonuclease activity	Positive
Hotstart (SYBR® Green qPCR)	No detectable amplification without a heat activation step. Detectable amplification with an activation step for 10 min at 95°C.
Residual *E.coli* DNA	< 1fg / *Taq* Unit
Bioburden	≤10 CFU/ml

8. Unit definition

One unit is defined as the amount of enzyme that incorporates, after activation step, 10 nmoles of dNTPs into acid insoluble form in 30 minutes at 74 °C.

Reaction Conditions

For a 100 µl Reaction

HGS Diamond *Taq*® Reaction Buffer (10x)	10 µl
$MgCl_2$ solution	6 µl (1.5 mM)
HGS Diamond *Taq*®	0.8 to 2.5 units
dNTP	200 µM each dNTP
Primers	0.1 nmol each
H_2O	As required
DNA template	As required

Magnesium

This DNA polymerase is a magnesium-dependent enzyme. We recommend increasing the magnesium concentration for long DNA fragments. Excess Mg^{2+} stabilizes the DNA double strand and consequently prevents complete denaturation of DNA, which reduces the extension yield. It may also stabilize spurious primer/template annealing, thus decreasing specificity.

Recommendation

Homogenize HGS Diamond Taq® solution by flipping the tube 4 to 5 times.

Cycling conditions

Classical PCR protocol used for 500 bp lambda DNA amplification*

	95°C	10 min
	(enzyme activation + DNA denaturation)	
25 cycles	94°C	30 sec
	Tm –2°C	30 sec
	72°C	1 min/kb
	72°C	7 min
	4°C	end temperature

**Condition will vary from reaction to reaction and may need optimization for maximal performances. Duration and temperature for denaturation and annealing steps depend on the type of cycler and primers design.*

Disclaimer

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