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Red Diamond *Taq*® DNA Polymerase

Specification Sheet

Reference: TAQ-I041

Eurogentec products are sold for research or laboratory use only and are not to be administrated to humans or used for medical diagnostics.

Source

Red Diamond *Taq*® is a highly thermostable enzyme produced and purified from recombinant *Escherichia coli* bacterium containing the *Thermus aquaticus* DNA Polymerase gene. This thermophilic eubacterium strain lacks *Taq* I restriction endonuclease.

Intended use

The enzyme shows very good fidelity and catalyzes 5'→3' synthesis of DNA with no detectable 3'→5' exonuclease activity. The enzyme has the "extendase" activity allowing TA cloning. This enzyme corresponds to the Diamond *Taq*® with a red dye allowing visual confirmation of pipetting. Red Diamond *Taq*® is particularly suited for PCR & qPCR applications that require high sensitivity and ultra low levels of bacterial & fungal DNA and/or visual confirmation. 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 Red Diamond *Taq*® reaction buffer (10 X)*	Volume 25 mM $MgCl_2$
TAQ-I041-100 (sample)	100	20 µl	5 U/µl	1 ml	1 ml
TAQ-I041-1000	1000	200 µl	5 U/µl	6 ml	6 ml
TAQ-I041-5000	5000	1 ml	5 U/µl	30 ml	30 ml
TAQ-I041-25000	5 x 5000	5 x 1 ml	5 U/µl	5 x 30 ml	5 x 30 ml

*750 mM Tris-HCl pH 8.8 (at 19°C), 200 mM $(NH_4)_2SO_4$, 0.1% (v/v) Tween 20 and stabilizer.

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), red dye 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	Red solution
Identity (SDS-PAGE)	MW approx. 95 kDa
Volume activity	> 5 U/µl
Purity (SDS-PAGE)	> 98%
Performance test: PCR on λ DNA	0.5 kb fragment positive down to 5 pg
Performance test: PCR on genomic DNA	0.1 kb fragment positive down to 10 pg
Ribonucleases (up to 10 U, 1h, 37 °C)	Not detectable
Endonucleases (up to 30 U, 16h, 65 °C)	Not detectable
Exonucleases (up to 30 U, 16h, 65 °C)	Not detectable
Nicking activity (up to 30 U, 16h, 65 °C)	Not detectable
***E. coli* residual DNA**	< 1 fg / Taq Unit
Bioburden	≤ 10 CFU/ml
Stability	24 months (at -20°C) from date of manufacture
Animal-derived additives	None

Unit definition

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

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Reaction Conditions

For a 100 µl Reaction

Red Diamond *Taq*® Reaction Buffer (10x)	10 µl
$MgCl_2$ solution	6 µl (1.5 mM)
Red Diamond *Taq*®	0.8 to 2.5 units
dNTP	200 µM each dNTP
Primers	As required
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 Mg2+ 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 Red Diamond Taq® solution by flipping the tube 4 to 5 times.

10. Cycling conditions

Classical PCR protocol used for 500 bp lambda DNA amplification*

		95°C	10 min (enzyme activation + DNA denaturation)
		94°C	30 sec
25 cycles		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. We advise you to check primer design by using primer design software.

Disclaimer

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