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**SilverStar DNA polymerase****ME-0074-05 • ME-0074-SA**

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

Source and Description

SilverStar DNA Polymerase is a high-performance thermostable DNA polymerase purified from *Thermus aquaticus* (1). SilverStar offers consistent results across a wide range of assays. SilverStar leaves an A' overhang such that the primer extension product is suitable for effective integration into TA cloning vectors.

Quality control

Each lot is tested for the absence of nicking and priming activities, exonucleases and non-specific endonucleases.

Shipping conditions

Product is shipped at +4°C. However, due to high stability features, trial samples are shipped at room temperature and we recommend usage of trial sample within the following 3-4 weeks.

Storage conditions

SilverStar DNA Polymerase can be stored at -20°C, in a constant temperature freezer for 12 months (1). SilverStar will remain stable if stored as specified.

Repeated freeze-thaw cycles will affect the stability of Reaction Buffer. The Buffer will remain stable at +4°C for a minimum of one month.

Package contents

Reagent	Volume	Units	Concentration	Description
SilverStar DNA polymerase	100 µl 5 µl	500 U 25 U	5 U/µl 5 U/µl	DNA polymerase ME-0074-05 DNA polymerase ME-0074-SA
10x reaction buffer (yellow cap)	2x 1.5 ml (ME-0074-05) 1x 1.5 ml (ME-0074-SA)		10 x	160mM (NH ₄) ₂ SO ₄ , 670mM Tris-HCl (pH 8.8 at 25°C), 0.1% Tween-20. Without MgCl ₂
MgCl₂	2x 1.5 ml (ME-0074-05) 1x 1.5 ml (ME-0074-SA)		50 mM	MgCl ₂ stock solution

Analysis conditions

25 mM TAPS, pH 9.3 (at 25°C); 50 mM KCl; 2 mM MgCl₂; 1 mM β-mercaptoethanol; 250 µM each dCTP, dGTP, dTTP; 250 µM (³H) dATP (0.05 Ci/mmol); activated salmon sperm DNA (1.25 µg/µl); total volume of 50 µl.

Associated activities

Endonuclease and exonuclease activities were not detectable after 2 and 1-hour incubations, respectively, of 1 µg lambda DNA and 0.22 µg of EcoRI-digested lambda DNA at 72°C in the presence of 15-20 units of SilverStar DNA polymerase.

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 72°C.

Reaction Conditions**For a 50 µl volume**

10x Reaction Buffer	5 µl
50mM MgCl ₂ Solution	1.5 – 4.0 µl
dNTP final concentration	250 – 500 µM each dNTP
>20mM dNTP Mix (related product)	2.5 – 5 µl
Template and Primers	as required
Enzyme	0.5 – 1.0 µl
Water	up to 50 µl

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**Always keeping on ice**

MgCl₂ concentration: this polymerase is a magnesium-dependent enzyme. The supplied 50mM MgCl₂ solution should be used to adjust magnesium ion concentration. Excessive amount of 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.

Cycling conditions

Denaturation 94-96°C
 Annealing see below
 Elongation suggested 15-30 sec/kb at 70-72°C

Time and temperature for denaturation and annealing steps depend on the type of machine and primers. We advise that you check primer design using primer design software.

This data is intended for use as a guide only; conditions will vary from reaction to reaction and may need optimisation.

Troubleshooting guide

Observation	Recommended solution(s)
No or low yield of extended product	Enzyme concentration too low -increase the amount enzyme in 0.5 U increments Magnesium concentration too low – increase concentration in 0.25 mM increments. Primer annealing too low – Increase annealing temperature
Appearance of a smear	Reduce the quantity of polymerase. This should not exceed 1 U/50 µl, except for long fragments or not highly purified DNA
Appearance of contaminating bands	Reduce the magnesium concentration to 1.5 mM, or use Hot start technique

Related product

Reagent	Package size	Reference
dNTP Mix 20 mM total	1 X 20 µmol 5 X 20 µmol 10 X 20 µmol	NU-0010-10 NU-0010-50 NU-0010-100
dNTP Set 5mM each NTP	4 X 5 µmol 4 X 25 µmol	NU-0020-10 NU-0020-50
HotGoldStar DNA Pol	500 units	ME-0073-05

For further information please contact our Customer Help Desk:**For Europe:**

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