For research use only

ISO9001

rTaq DNA Polymerase

Product Description	Quantity	Cat. No.	Remarks
rTaq DNA Polymerase	500 unit	EBT-1013	5 unit/μl
	500 unit	EBT-1014	with 1 ml dNTP mix

Description

Recombinant rTaq DNA polymerase is overexpressed and purified from an *E.coli* strain containing the gene of *Thermus aquaticus* DNA polymerase. rTaq DNA polymerase is modified to increase thermostability and performance in conventional PCR process.

Storage Buffer

5 unit/μl in 20 mM Tris-HCl, pH8.0, 100 mM KCl, 0.1 mM EDTA, 1 mM DTT, 0.5% Tween 20, and 50% Glycerol.

Unit Definition

One unit is defined as the amount of enzyme required to catalyze the incorporation of 10 nmole of dNTP into an acid-insoluble form in 30 min at 72℃.The reaction conditions are : 25 mM TAPS, pH9.3, 50 mM KCl, 2 mM $MgCl_2$, 1 mM β-mercaptoethanol, 200 μM each dNTPs, 100 μM [α-^{32}P]dCTP, 12.5 μg activated calf thymus DNA in a total volume of 50 μl.

10x PCR Reaction Buffer w/ $MgCl_2$

500 mM Tris-HCl, pH9.0, 160 mM $(NH_4)_2SO_4$, 25 mM $MgCl_2$, 1% Tween 20, 1 mg/ml BSA.

QC tests

Activity, SDS-PAGE purity, performance tests.

Storage Condition

Store at -20℃.

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Standard Protocol

1. Prepare 50 μl PCR solution as follows:

PCR grade distilled water :	- μl
10x reaction buffer :	5 μl
10 mM dNTP mix (2.5 mM each) :	4 μl
Primer (10 pmol/μl) :	1 μl each
Template :	1-10 ng
rTaq DNA Polymerase :	0.5 μl (2.5 unit)

Adjust final vol. to 50 μl with PCR grade distilled water

*Note : Always, rTaq DNA polymerase should be added last to the mixture

2. Set PCR cycling as follows :

Initial denature at 95°C : 3 min

		<1 kbp	1-4 kpb
Denature	95°C	30 sec	30 sec
Anneal	Tm-4°C	30 sec	30 sec
Extend	72°C	45 sec	30-60 sec/1 kbp

25-40 PCR cycles

Trouble-Shooting

1. No products
   - Confirm your template is intact : Try another reaction with a result assured primer pair and templates
   - Be sure all the component are correctly added and working well : Sometimes low graded dNTP may inhibit the reaction, and degraded primers can result in low sized PCR fragments
2. Smear bands or smeared background
   - Reduce template concentration : High concentration of template can lead to smearing of PCR products. Generally, 1-10 ng of plasmid DNA is working well
   - Increase annealing temperature
3. Non-specific bands
   - Increase annealing temperature
   - Consider using PCR additives, like 1-2% DMSO and 0.5-1x Q buffer
   - Confirm specificity of your primers
4. Low yield
   - Increase enzyme concentration in the reaction
   - Increase PCR cycle number
   - Be sure appropriate concentration of your template is added