Direct Blood *Eco*Taq DNA Polymerase, 5U/µl

100U, 500U

Cat No: EBP100, EBP500

Shipping : Ship with blue ice.
Storage : Store at -20°C. Avoid freeze and thaw cycles.

General Information

Direct Blood *Eco*Taq DNA Polymerase is an engineered variant of Taq DNA polymerase, developed through N-terminal truncation and targeted mutations. This modification enhances tolerance to common PCR inhibitors, enabling direct DNA amplification from whole blood samples of human and mouse origin without the need for prior DNA extraction or purification.

Despite the relatively low DNA content and complexity of certain sample types, the enzyme is also suitable for direct DNA amplification from saliva, serum, and urine samples without the need for prior DNA extraction or purification.

The enzyme generates PCR products with a 3′-A overhang, allowing direct use in T/A cloning applications.

Direct Blood *Eco*Taq DNA Polymerase is supplied together with 10x Reaction Buffer. 10×PCR Buffer contains 30 mM $MgCl_2$.

Protocol

1. Gently invert the Direct Blood *Eco*Taq DNA Polymerase several times to ensure thorough mixing before use.

2. Place the PCR thin-walled tube on ice and add all components except the whole blood sample. Prepare the following reaction mixture.

PCR Setup

Component	Amount
Direct Blood *Eco*Taq DNA Polymerase, 5U/µl	1 µL
10x Reaction Buffer	5 µl
Forward primer, 10 µM	2 µl
Reverse primer, 10 µM	2 µl
dNTP Mix, 10 mM each	1 µl
Whole Blood*	<10%
ddH_2O	up to 50 µl
Total	50 µl

* Prior to adding the whole blood sample, ensure it is thoroughly mixed by pipetting up and down. Whole blood samples may be collected using anticoagulants such as sodium heparin, Na-EDTA, K-EDTA, or sodium citrate. A final blood concentration of 5–10% in the reaction is generally recommended, as higher concentrations may inhibit amplification. For templates with high GC content, the addition of 10% DMSO is recommended.

Important Note: PCR reaction conditions might need optimization with additional $MgCl_2$ (not provided) depending on the primer binding properties.

3. Add the whole blood to the bottom of the tube.

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4. Run the reaction using the following conditions.

Temperature	Time	Cycles
95°C	5 min	
95°C	30 sec	
50-68°C	30 sec*	35-40
72°C	250-500bp/min	
72°C	10 min	
4°C	∞	

*The annealing step is typically performed for 30 seconds to 1 minute. The temperature should be optimized based on primer characteristics, for example by gradient PCR.

Important Notes

Preheat the thermal cycler to 94–95 °C, then place the samples into the instrument and initiate the cycling program.

Direct Blood *Eco*Taq DNA Polymerase exhibits reduced cold sensitivity and partial hot-start characteristics. Reaction components can be prepared on ice, and non-specific amplification can be minimized by adding the polymerase at last step and preheating the thermocycler to the denaturation temperature (95°C).

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