

Advance™ DNA Polymerase

(Cat# M1151-250, -1000; 5 U/µl; Store at -20°C)

I. Introduction:

BioVision's AdvanceTM Taq DNA Polymerase is a novel DNA polymerase with strategically engineered mutations resulting in a robust, high fidelity polymerase. AdvanceTM Taq DNA Polymerase has exceptional 3' to 5' exonuclease activity that endows it with superior accuracy over competitor polymerases. This novel enzyme has intrinsically high processivity and is engineered to have an improved binding affinity for DNA resulting in highly successful PCR. The AdvanceTM 2X PCR sophisticated buffer system not only tolerates high AT and GC content, but also many PCR inhibitors commonly found in a typical DNA sample. BioVision's AdvanceTM Taq DNA Polymerase is the ultimate solution for a robust PCR system with high fidelity and yield.

II. Application:

- · Routine PCR amplification
- PCR success with AT and GC Rich sequences
- High-throughput PCR
- RACE
- · NGS Library construction

III. Key Features:

- · High fidelity PCR
- Robust PCR performance, resistant to most PCR inhibitors commonly found in samples (including plant samples)
- PCR success with A/T and G/C rich templates
- Highly processive, for high yield amplification

IV. Unit Definition:

One unit of the enzyme catalyzes the incorporation of 10 nmol of deoxy ribonucleotides into a polynucleotide fraction in 30 mins at 74°C.

V. Package Contents:

Cat. No.	M1151-250	M1151-1000	Part Number
Concentration	250 U	1000 U	-
Advance [™] DNA Polymerase (5 U/µI)	50 µl	200 µl	M1151-XX-1
2X PCR Buffer, with Mg ²⁺	2 x 1 ml	6 x 1 ml	M1151-XX-2
25 mM MgSO ₄	1 ml	1 ml	M1151-XX-3

VI. Storage Buffer Components:

50 mM Tris-HCl (pH 8.0), 100 mM NaCl, 0.1 mM EDTA, 5 mM DTT, 50% glycerol and 1.0% Triton X-100.

VII. Shipment and Storage:

Upon arrival, Advance[™] Taq DNA Polymerase should be stored at -20°C. Avoid repeated freeze-thaw cycles of all the Advance[™] Taq components to retain maximum performance. All Advance[™] Taq components are stable for 1 year from the date of shipping if stored and handled properly. Briefly centrifuge small vials prior to opening.

VIII. PCR Protocol:

- The following basic protocol serves as a general guideline and starting point for any PCR amplification. Optimal reaction conditions (incubation times, temperatures, concentration of Taq DNA Polymerase, primers, MgSO4 and template DNA) may vary and need to be optimized for each specific PCR.
- All PCR experiments should be assembled in a nuclease-free environment. In addition, DNA sample preparation, reaction set-up and subsequent reaction(s) should be performed in separate areas to avoid cross contamination.
- A negative control reaction (omitting template DNA) should always be performed in tandem with sample PCR to confirm the absence
 of DNA contamination.
- 1. Add the following components to a sterile 0.2 ml PCR tube sitting on ice.

Components	Volume	Final concentration
Advance™ Taq DNA Polymerase (5 U/µI)	1 µl	5 U
10 μM Forward Primer	1-2.5 µl	200-500 nM
10 μM Reverse Primer	1-2.5 µl	200-500 nM
Template DNA	~100 ng	~2 ng/µl
2X PCR buffer, with Mg ²⁺	25 µl	1X
25 mM MgSO ₄ (optional)*	0-3 μΙ	1.5-3 mM
dNTP Mix (10 mM)	1 µl	200 μΜ
Water, Nuclease-free	Up to 50 µl	-

^{*}Optimal Mg^{2+} concentration is specific to each DNA template-primer set and can only be determined experimentally.

3. Incubate tube in a thermal cycler at 94°C for 3 mins to completely denature the template.

4. Perform 30-35 cycles of PCR amplification as follows:

Denature: 94°C for 30 sec; Anneal: 45 - 72°C for 30 sec; Extend: 72°C for 1 min/1 kb template

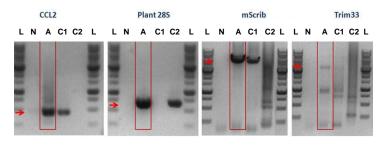
- 5. Incubate for an additional 5 mins at 72°C and maintain the reaction at 4°C. The samples can be stored at -20°C until use.
- Analyze the amplified products by agarose gel electrophoresis and visualize by ethidium bromide staining. Use appropriate molecular weight standards.

^{*}We recommend preparing a master mix for multiple reactions to minimize reagent loss and enable accurate pipetting.

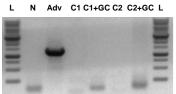
^{2.} Mix contents of tube and centrifuge briefly.



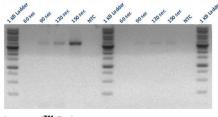
IX. Sensitivity:



Advance[™] Polymerase's Robustness: Selected difficult templates including a plant gDNA sample were amplified using Advance (A) and Competitor's DNA polymerase (C1 & C2); L: Ladder; N: Neg Ctrl.



High GC tolerance of Advance™ Taq DNA Polymerase: L:
Ladder; N: Neg ctrl; Adv: Advance™ DNA Polymerase; C1: Competitor1; C1+GC:
Competitor1+GC enhancer; C2: Competitor2; C2+GC: Competitor2+GC enhancer.



Speed of AdvancedTM Polymerase: A 3.5 kb target was amplified with various extension time using AdvanceTM or ReadyTM Taq DNA polymerase.

X. Related Products:

BV product name	BV Cat. No.	
Ready [™] DNA Polymerase	M1146-1000	
Ready [™] DNA Polymerase	M1146-5000	
Ready [™] DNA Polymerase	M1146-10,000	
Robust Ready [™] DNA Polymerase	M1147-250	
Robust Ready [™] DNA Polymerase	M1147-1000	
Breeze [™] DNA Polymerase	M1148-250	
Breeze [™] DNA Polymerase	M1148-1000	
Fire Start [™] DNA Polymerase	M1149-250	
Fire Start [™] DNA Polymerase	M1149-1000	
Distant [™] DNA Polymerase	M1150-250	
Distant [™] DNA Polymerase	M1150-1000	
Advance [™] DNA Polymerase	M1151-250	
Advance [™] DNA Polymerase	M1151-1000	
Outstretched [™] DNA Polymerase	M1152-250	
Outstretched [™] DNA Polymerase	M1152-1000	
Blood Advance [™] DNA Polymerase	M1153-100	
Blood Advance [™] DNA Polymerase	M1153-400	
Image Green™ 100 bp DNA Marker	M1155-500	
Image Green™ 1 kb DNA Marker	M1156-500	

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