



## ProFoldin

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# INSTRUCTIONS

## ProFoldin

### Human DNA Polymerase Beta Assay Kits

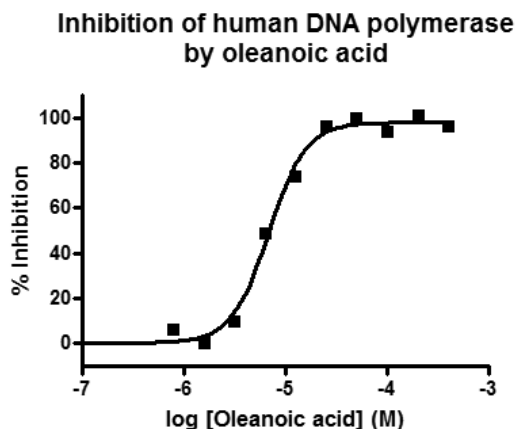
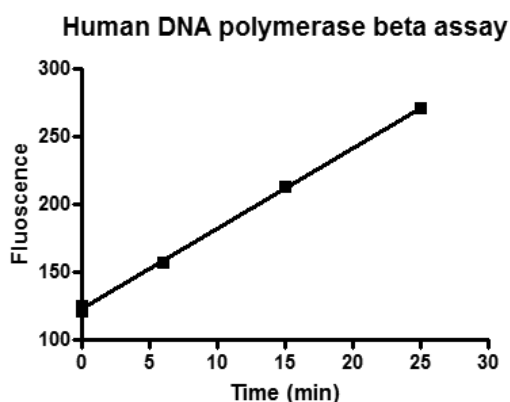
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**Human DNA Polymerase Beta Assay Kit**  
**Human DNA Polymerase Beta Assay Kit Plus**

**Catalog No: DPB100K**  
**Catalog No: DPB100KE**

#### Introduction

DNA polymerase beta play key roles in the base excision repair (BER) process. It fills the small gaps (1 – 6 bases) of one DNA stand using the complementary strand as a template. Human DNA polymerase beta is an anti-cancer target. The human DNA polymerase beta assay is based on detection of the DNA polymerase activity in the presence of a DNA template and dNTPs. The Assay Kit (catalog number DPB100K) is to measure the activity of DNA polymerase beta and test DNA polymerase beta inhibitors.



Each **Human DNA Polymerase Beta Assay Kit (Catalog No: DPB100K)** includes all the assay kit components except the enzyme for 100 assays in a 384-well plate assay format: 400  $\mu$ l of 10 x Buffer, 33  $\mu$ l of 100 x DNA template (3  $\mu$ M), 33  $\mu$ l of 100 x dNTP mix (10 mM), 1550  $\mu$ l of 2 x Dye, 1550  $\mu$ l of 50 mM EDTA.

Each **Human DNA Polymerase Beta assay Kit Plus (Catalog No: DPB100KE)** includes all the assay kit components for 100 assays in 384-well plate assay format: 400  $\mu$ l of 10 x Buffer, 33  $\mu$ l of 100 x DNA template (3  $\mu$ M), 33  $\mu$ l of 100 x dNTP mix (10 mM), 33  $\mu$ l of 100 x human DNA polymerase beta, 1550  $\mu$ l of 2 x Dye, 1550  $\mu$ l of 50 mM EDTA.

#### Assay protocol

The following assay protocol is based on the 384-well plate assay format (plate type: Matrix 4318 or alike). The reaction volume is 30  $\mu$ l and the final assay volume is 60  $\mu$ l. For 96-well plate assays (plate type: Costar 3915 or alike), the reaction volume is 50  $\mu$ l and the final assay volume is 100  $\mu$ l.

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## 1. Reagent preparation:

- (1) 10 x DNA: Dilute the 100 x DNA 10-fold with water. Each assay uses 3  $\mu$ l of 10 x DNA.
- (2) 10 x enzyme: Dilute the 100 x Human DNA polymerase beta 10-fold with the 1 x assay buffer.  
Each assay uses 3  $\mu$ l of 10 x enzyme.
- (3) 10 x dNTP mix: Dilute the 100 x dNTP mix 10-fold with water. Each assay uses 3  $\mu$ l of 10 x dNTP mix.
- (4) 1 x dye: Dilute the 2 x fluorescence dye 2-fold with 50 mM EDTA. Each assay uses 30  $\mu$ l of 1 x dye.

## 2. Reaction:

The total volume of each reaction mixture is 30  $\mu$ l including 18  $\mu$ l of H<sub>2</sub>O, 3  $\mu$ l of 10 x Buffer, 3  $\mu$ l of 10 x DNA template, 3  $\mu$ l of 10 x enzyme, 3  $\mu$ l of 10 x dNTP mix. Incubate the reaction mixture at 37°C for 60 min.

## 3. Detection:

Mix 30  $\mu$ l of the 1 x fluorescence dye with 30  $\mu$ l of the reaction mixture. Measure the fluorescence intensity at 535 nm using the excitation wavelength at 485 nm.

## Assay Protocol for enzyme inhibition

The assay can be optimized in terms of assay window, assay linearity and sensitivity to competitive inhibitors. ProFoldin offers HTS assay development service. For more information, please visit our website at <http://www.profoldin.com/services.html>.

## Related Products

DPA100KE	<i>E. coli</i> DNA Polymerase III Alpha Assay Kit Plus
HDPA100K	Human DNA Polymerase Alpha Assay Kit
DPG100K	Human DNA Polymerase Gamma Assay Kit
DPG100KE	Human DNA Polymerase Gamma Assay Kit Plus
RPA100KE	<i>E. coli</i> RNA Polymerase Assay Kit Plus
RPA100KSE	<i>S. aureus</i> RNA Polymerase Assay Kit Plus
T7RPA100KE	T7 RNA Polymerase Assay Kit Plus
MRPA100K	Human Mitochondrial RNA Polymerase Assay Kit
AMV100KE	AMV Reverse Transcriptase Assay Kit Plus
HIV100KE	HIV Reverse Transcriptase Assay Kit Plus
MLV100KE	M-MLV Reverse Transcriptase Assay Kit Plus
VRT100K	Viral RNA-dependent RNA Polymerase Assay Kit - 100
HCMV100K	Human Cytomegalovirus (HCMV) DNA Polymerase Assay Kit
PNP100KE	Polynucleotide Phosphorylase (PNPase) Assay Kit Plus

For more information of drug targets and enzyme assays, please visit [www.profoldin.com](http://www.profoldin.com).