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INSTRUCTIONS

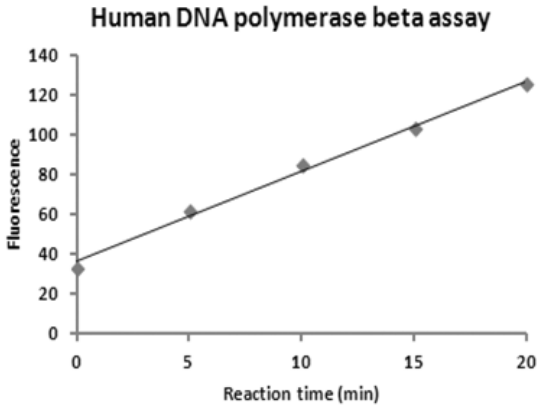
ProFoldin
Human DNA Polymerase Beta Assay Kits

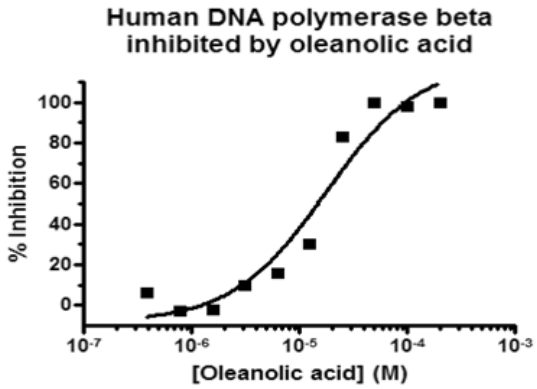
Human DNA Polymerase Beta Assay Kit **Catalog No: DPB100K**

Human DNA Polymerase Beta Assay Kit Plus **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 the principle that the repaired DNA forms more stable DNA duplex than the un-repaired one. The Assay Kit (catalog number DPB100K) is to measure the formation of the repaired DNA by its fluorescence signal in the presence of a fluorescence dye.





Each **Human DNA Polymerase Beta Assay Kit (Catalog No: DPB100K)** includes 800 µl of 10 x Buffer BP, 55 µl of 100x DNA template, 55 µl of 100 x dNTP mix, 22 ml of Reagent U and 420 µl of 10 x fluorescence dye for 100 assays of human DNA polymerase reactions in a 96-well plate format. The kit does not include human DNA polymerase beta.

Each **Human DNA Polymerase Beta assay Kit Plus (Catalog No: DPB100KE)** includes 800 µl of 10 x Buffer BP, 55 µl of 100x DNA template, 55 µl of 100 x dNTP mix, 55 µl of 100 x human DNA polymerase beta, 22 ml of Reagent U and 420 µl of 10 x fluorescence dye for 100 assays of human DNA polymerase reactions in a 96-well plate format.

Assay Protocol

1. **Reagent preparation:**

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10 x DNA: dilute the 100 x DNA 10-fold with water
10 x dNTP mix: dilute the 100 x dNTP 10-fold with water
Reagent U: provided with the kit
1 x Fluorescence dye: dilute the 10 x fluorescence dye 10-fold with water

2. **Reaction**:
The total volume of each reaction mixture is 50 µl including: 30 µl of H_2O, 5 µl of 10 x buffer (Buffer PB), 5 µl of 10 x DNA, 5 µl of 10 x human DNA polymerase beta, 5 µl of 10 x dNTP mix. Incubate the reaction mixture in a standard black 96-well plate at room temperature for 30 min to 60 min.

Note: The final concentrations in the assay reaction are 30 mM Tris-HCl, pH 8.0, 10 mM $MgCl_2$, 1 mM DTT, 5 % glycerol, 100 nM DNA, 0.1 mM dATP, 0.1 mM dGTP. The recommended final enzyme concentration in the reaction mixture is 2 U/ml or 2 nM.

3. **Detection**:
Add 200 µl of Reagent U into the 50 µl of reaction mixture. Then add 40 µl of the 1 x fluorescence dye. Mix the reaction solution and 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	*E. coli* 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	*E. coli* RNA Polymerase Assay Kit Plus
RPA100KSE	*S. aureus* RNA Polymerase Assay Kit Plus
T7RPA100KE	T7 RNA Polymerase Assay Kit Plus
MRPA100K	Human Mitochondrial RNA Polymerase Assay Kit

More information of drug targets and enzyme assays

For more information of drug targets and enzyme assays, please visit www.profoldin.com or send emails to info@profoldin.com.