APA988Mu01 100μg
Active Cytochrome P450 2E1 (CYP2E1)
Organism Species: *Mus musculus (Mouse)*
Instruction manual

FOR RESEARCH USE ONLY
NOT FOR USE IN CLINICAL DIAGNOSTIC PROCEDURES

13th Edition (Revised in Aug, 2023)

[PROPERTIES]

Source: Prokaryotic expression.
Host: *E. coli*
Residues: Phe378~Ser493
Tags: N-terminal His-tag
Purity: >95%
Endotoxin Level: <1.0EU per 1μg (determined by the LAL method).
Buffer Formulation: PBS, pH7.4, containing 0.01% SKL, 5%Trehalose .
Original Concentration: 200μg/mL
Applications: Cell culture; Activity Assays.
(May be suitable for use in other assays to be determined by the end user.)
Predicted isoelectric point: 6.7
Predicted Molecular Mass: 14.4kDa
Accurate Molecular Mass: 14kDa as determined by SDS-PAGE reducing conditions.

[USAGE]

Reconstitute in 10mM PBS (pH7.4) to a concentration of 0.1-1.0 mg/mL. Do not vortex.

[STORAGE AND STABILITY]

Storage: Avoid repeated freeze/thaw cycles.
Store at 2-8ºC for one month.
Aliquot and store at -80ºC for 12 months.

Stability Test: The thermal stability is described by the loss rate. The loss rate was determined by accelerated thermal degradation test, that is, incubate the protein at 37ºC for 48h, and no obvious degradation and precipitation were observed. The loss rate is less than 5% within the expiration date under appropriate storage condition.

[SEQUENCE]

FRG YVIPKGTVVI PTLDSLLFDN
YEFPDPETFK PEHFLNENGK FKYSDYFKAF SAGKRVCVGE GLARMELFLL
LSAILQHFNL KSLVDPKDID LSPVTIGFGS IPREFKLCVI PRS

[ACTIVITY]

The cytochrome P450 enzyme CYP2E1 catalyzes the oxidative metabolism of many solvents and other small organic molecules. CYP2E1 is expressed in adult and fetal human liver in addition to extrahepatic tissues such as lung and placenta. Treatment of primary cultures of human hepatocytes with ethanol induces CYP2E1 protein, and this is consistent with the finding that hepatic CYP2E1 protein and mRNA levels are increased in individuals with alcoholism. Although only a few drugs (e.g., acetaminophen have been identified as substrates for CYP2E1, many low molecular weight procarcinogens are activated by this cytochrome P450 (P450). Chlorzoxazone 6-hydroxylation, N-nitrosodimethylamine N-demethylation and p-nitrophenol hydroxylation can be used to measure the catalytic activity of CYP2E1. Thus, the recombinant mouse CYP2E1 activity was measured by its ability to hydroxylate p-nitrophenol to p-nitrocatechol. The reaction was performed in 50 mM potassium phosphate, pH 7.4 (Assay Buffer), initiated by addition 20 μL of 500 ug/ml CYP2E1 to 10 μL of 5 mM substrate p-nitrophenol and 30 ul of 26 mM NADPH in a total volume of 500 ul. Incubated at 37 ℃ for 30min, then read at a wavelength of 535 nm after acidification of the reaction mixture with trichloroacetic acid followed by neutralization using 2 M NaOH.

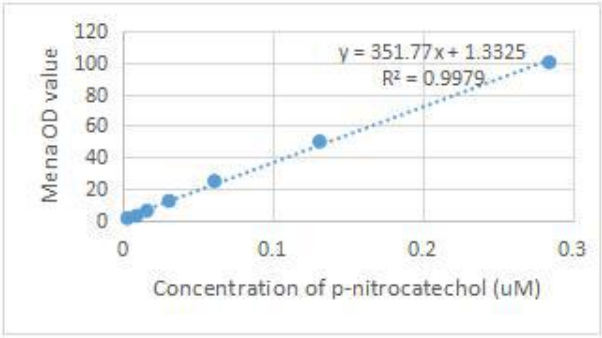

Figure 1. The standard curve of p-nitrocatechol

OD535nm	p-nitrocatechol (product) uM
0.2847	100
0.1315	50
0.0614	25
0.031	12.5
0.0163	6.25
0.0094	3.125
0.0033	1.5625

One unit of enzyme activity is defined as the 1 μg of enzyme required to convert 1 pmol of p-nitrophenol to p-nitrocatechol in 1 min at 37°C. The specific activity of recombinant mouse CYP2E1 is 3.6 pmol/min/μg.

Specific Activity (pmol/min/μg)= $\frac{\Delta OD * F}{T * N}$

△OD=Adjusted for Substrate Blank

F=Conversion Factor(convert from standard curve of p-nitrocatechol)

T= Time

N=Amount of enzyme

[IDENTIFICATION]

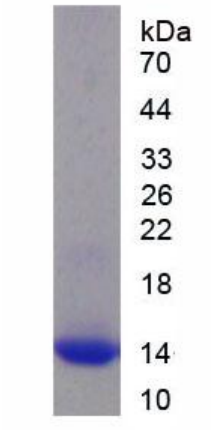

Figure 2. SDS-PAGE

Sample: Active recombinant CYP2E1, Mouse

[IMPORTANT NOTE]

The kit is designed for research use only, we will not be responsible for any issue if the kit was used in clinical diagnostic or any other procedures.