APC374Hu01 50μg
Active Carboxylesterase 1 (CES1)
Organism Species: *Homo sapiens* (Human)
Instruction manual

FOR RESEARCH USE ONLY
NOT FOR USE IN CLINICAL DIAGNOSTIC PROCEDURES

1st Edition (Apr, 2016)

[PROPERTIES]

Source: Prokaryotic expression.
Host: *E. coli*
Residues: Gly18~Leu299
Tags: N-terminal His-tag
Purity: >90%
Endotoxin Level: <1.0EU per 1μg (determined by the LAL method).
Buffer Formulation: 20mM Tris, 150mM NaCl, pH8.0, containing 0.01% sarcosyl and 5% trehalose.
Original Concentration: 400μg/mL
Applications: Cell culture; Activity Assays.
(May be suitable for use in other assays to be determined by the end user.)
Predicted isoelectric point: 6.8
Predicted Molecular Mass: 34.0kDa
Accurate Molecular Mass: 35kDa as determined by SDS-PAGE reducing conditions.

[USAGE]

Reconstitute in 20mM Tris, 150mM NaCl (pH8.0) to a concentration of 0.1-1.0 mg/mL. Do not vortex.

[STORAGE AND STABILITY]

Storage: Avoid repeated freeze/thaw cycles.
Store at 2-8^{o}C for one month.

Aliquot and store at -80°C for 12 months.

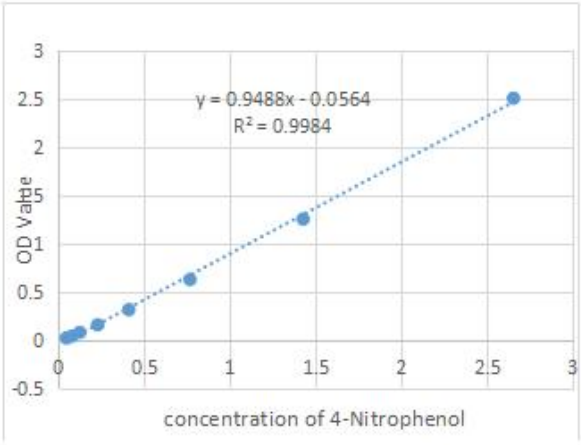
Stability Test: The thermal stability is described by the loss rate. The loss rate was determined by accelerated thermal degradation test, that is, incubate the protein at 37°C for 48h, and no obvious degradation and precipitation were observed. The loss rate is less than 5% within the expiration date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

[SEQUENCE]

GHP SSPPVVDTVH GKVLGKFVSL EGFAQPVAIF
LGIPFAKPPL GPLRFTPPQP AEPWSFVKNA TSYPPMCTQD PKAGQLLSEL
FTNRKENIPL KLSEDCLYLN IYTPADLTKK NRLPVMVWIH GGGLMVGAAS
TYDGLALAAH ENVVVVTIQY RLGIWGFFST GDEHSRGNWG HLDQVAALRW
VQDNIASFGG NPGSVTIFGE SAGGESVSVL VLSPLAKNLF HRAISESGVA
LTSVLVKKGD VKPLAEQIAI TAGCKTTTSA VMVHCLRQKT EEELLETTL

[ACTIVITY]

carboxylesterase 1 (CES1) also known as Liver carboxylesterase 1 is a serine esterase and member of a large multigene carboxylesterase family. The protein Involved in the detoxification of xenobiotics and in the activation of ester and amide prodrugs. Hydrolyzes aromatic and aliphatic esters, but has no catalytic activity toward amides or a fatty acyl-CoA ester. Hydrolyzes the methyl ester group of cocaine to form benzoylecgonine. Thus, the recombinant human CES1 activity was measured by its ability to hydrolyze 4-Nitrophenyl acetate (4-NPA) to 4-Nitrophenol. The reaction was performed in 50 mM Tris, pH 7.5(Assay Buffer), initiated by addition 50 µL of various concentrations of CES1 (dilute by Assay Buffer) to 50 µL of 2 mM Substrate 4-NPA(100 mM stock in Acetone, dilute by deionized water). Incubated at 37℃ for 10min, then read at a wavelength of 400 nm.



4-Nitrophenol (product)mM	OD400nm
0.01953125	0.045
0.0390625	0.076
0.078125	0.123
0.15625	0.227
0.3125	0.409
0.625	0.766
1.25	1.426
2.5	2.653

Figure 1. The standard curve of 4-Nitrophenol

One unit of enzyme activity is defined as the 1μg of enzyme required to convert 1pmol of 4-Nitrophenyl acetate to 4-Nitrophenol in 1min at 37°C. The specific activity of recombinant human CES1 is 600 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 4-Nitrophenol)

T= Time

N=Amount of enzyme

[IDENTIFICATION]

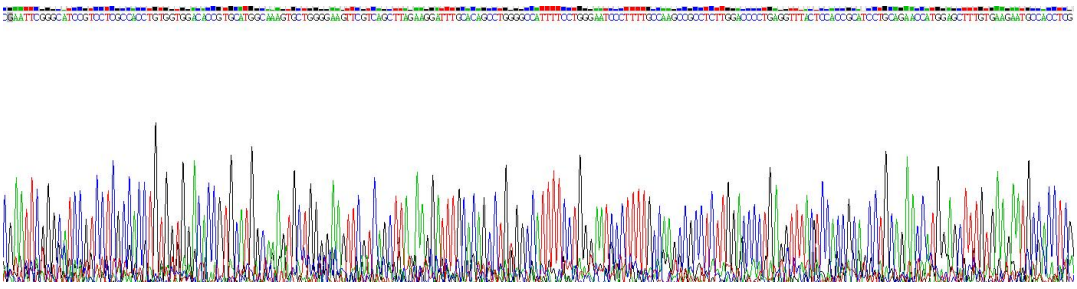


Figure 2. Gene Sequencing (extract)

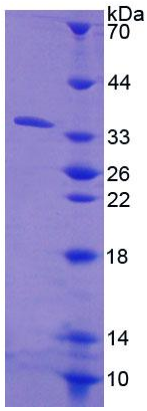


Figure 3. SDS-PAGE

Sample: Active recombinant human, CES1

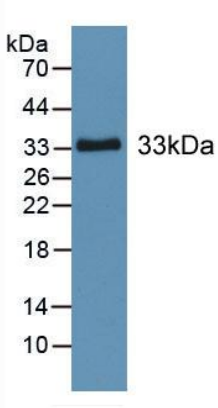


Figure 4. Western Blot

Sample: Recombinant human, CES1;

Antibody: Rabbit Anti-CES1 human Ab (PAC374Hu01)

[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.