

**APF283Hu01 100µg**

**Active Carboxylesterase 3 (CES3)**

**Organism Species: *Homo sapiens (Human)***

***Instruction manual***

FOR RESEARCH USE ONLY

NOT FOR USE IN CLINICAL DIAGNOSTIC PROCEDURES

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1st Edition (Apr, 2016)

## **[ PROPERTIES ]**

**Source:** Prokaryotic expression.

**Host:** *E. coli*

**Residues:** Ser398~Gln568

**Tags:** N-terminal His-tag

**Purity:** >94%

**Endotoxin Level:** <1.0EU per 1µg (determined by the LAL method).

**Buffer Formulation:** PBS, pH7.4, containing 0.01% SKL, 5% Trehalose.

**Applications:** Cell culture; Activity Assays.

(May be suitable for use in other assays to be determined by the end user.)

**Predicted isoelectric point:** 7.8

**Predicted Molecular Mass:** 23.4kDa

**Accurate Molecular Mass:** 23kDa 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 2-8°C for one month.

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

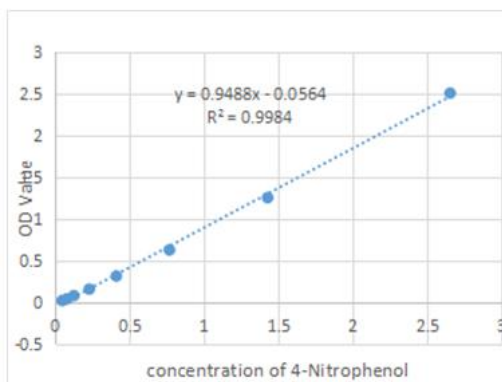
**Stability Test:** The thermal stability is described by the loss rate. The loss rate was determined by accelerated thermal degradation test, that is, incubate the protein at 37°C for 48h, and no obvious degradation and precipitation were observed. The loss rate is less than 5% within the expiration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

## **[ SEQUENCE ]**

SNS DAQAKCQAFQ EFMGDVFINV PTVSFSRYLR DSGSPVFFYE FQHRPSSFAK IKPAWVKADH  
GAEGAFVFGG PFLMDESSRL AFPEATEEEK QLSLTMMAQW THFARTGDPN SKALPPWPQF  
NQAQYLEIN PVPRAGQKFR EAWMQFWESET LPSKIQQWHQ KQKNRKAQ

## **[ ACTIVITY ]**

Carboxylesterase 3 (CES3) also known as Liver carboxylesterase 3 is a serine esterase and a member of a large family of carboxylesterases. 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 CES3 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 CES3 (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°C 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 1 $\mu$ g of enzyme required to convert 1pmol of 4-Nitrophenyl acetate to 4-Nitrophenol in 1min at 37°C. The specific activity of recombinant human CES3 is 700 pmol/min/ $\mu$ g.

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

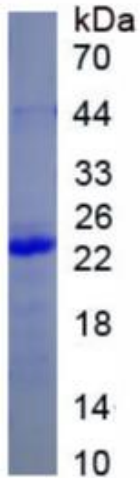
$\Delta$ OD=Adjusted for Substrate Blank

F=Conversion Factor(convert from standard curve of 4-Nitrophenol)

T= Time

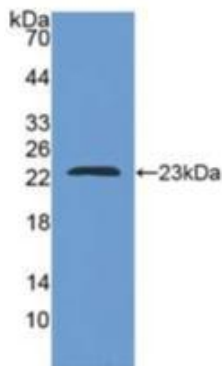
N=Amount of enzyme

## [ IDENTIFICATION ]



**Figure 2. SDS-PAGE**

Sample: Active recombinant CES3, Human



**Figure 3. Western Blot**

Sample: Recombinant CES3, Human;

Antibody: Rabbit Anti- Human CES3 Ab (PA F283Hu01)

**[ 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.