APF282Hu61 10μg Active Carboxylesterase 2 (CES2) Organism Species: *Homo sapiens (Human) Instruction manual*

FOR RESEARCH USE ONLY NOT FOR USE IN CLINICAL DIAGNOSTIC PROCEDURES

12th Edition (Revised in Aug, 2016)

[PROPERTIES]

Source: Eukaryotic expression. Host: 293F cell Residues: Gln27~Leu559 Tags: N-terminal His-tag Purity: >95% Endotoxin Level: <1.0EU per 1µg (determined by the LAL method). Buffer Formulation: PBS, pH7.4, containing 5% trehalose. Original Concentration: 600µg/mL Applications: Cell culture; Activity Assays. (May be suitable for use in other assays to be determined by the end user.) Predicted isoelectric point: 5.3 Predicted Molecular Mass: 60.6kDa Accurate Molecular Mass: 61kDa as determined by SDS-PAGE reducing conditions.

[<u>USAGE</u>]

Reconstitute in 10mM PBS (pH7.6) 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.

[<u>SEQUENCE</u>]

QDSASPIRTTHTGQVLGSLVHVKGANAGVQTFLGIPFAKPPLGPLRFAPPEPPESWSGVRDGTTHPAMCLQDLTAVESEFLSQFNMTFP SDSMSEDCLYLSIYTPAHSHEGSNLPVMVWIHGGALVFGMASLYDGSMLAALENVVVVIIQYRLGVLGFFSTGDKHATGNWGYLDQVAA LRWVQQNIAHFGGNPDRVTIFGESAGGTSVSSLVVSPISQGLFHGAIMESGVALLPGLIASSADVISTVVANLSACDQVDSEALVGCLR GKSKEEILAINKPFKMIPGVVDGVFLPRHPQELLASADFQPVPSIVGVNNNEFGWLIPKVMRIYDTQKEMDREASQAALQKMLTLLMLP PTFGDLLREEYIGDNGDPQTLQAQFQEMMADSMFVIPALQVAHFQCSRAPVYFYEFQHQPSWLKNIRPPHMKADHGDELPFVFRSFFGG NYIKFTEEEEQLSRKMMKYWANFARNGNPNGEGLPHWPLFDQEEQYLQLNLQPAVGRALKAHRLQFWKKALPQKIQELEEPEERHTEL

[ACTIVITY]

Carboxylesterase 2 (CES2) is a member of serine esterase family which can hydrolyze ester and amide bonds. 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 CES2 activity was measured by its ability to hydrolyze 4-Nitrophenyl acetate (4-NPA) to 4-Nitrophenol. The reaction was performed in 50 mM Tris, 150 mM NaCl, pH 7.5 (Assay Buffer), ainitiated by addition 50 μ L of various concentrations of CES2 (diluted by Assay Buffer) to 50 μ L of 2 mM Substrate 4-NPA (100 mM stock in Acetone, diluted by Assay Buffer). Incubated at 37°C for 10 min, then read at a wavelength of 400 nm.

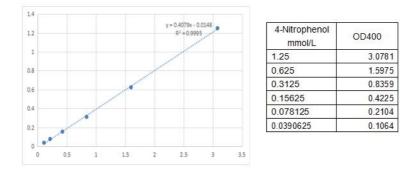


Figure 1. The standard curve of 4-Nitrophenol

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 CES2 is 502.86 umol/min/ μg .

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

 $\triangle \text{OD}\text{=}\text{Adjusted}$ for Substrate Blank

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

T= Time

N=Amount of enzyme

[IDENTIFICATION]

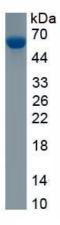


Figure 2. SDS-PAGE

Sample: Active recombinant CES2, Human

[<u>IMPORTANT NOTE</u>]

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.