

**APE824Hu01 100µg**  
**Active Aldehyde Dehydrogenase 1 Family, Member A1 (ALDH1A1)**  
**Organism Species: Homo sapiens (Human)**  
***Instruction manual***

FOR RESEARCH USE ONLY  
NOT FOR USE IN CLINICAL DIAGNOSTIC PROCEDURES

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13th Edition (Revised in Aug, 2023)

## **[ PROPERTIES ]**

**Source:** Prokaryotic expression.

**Host:** *E. coli*

**Residues:** Ser2~Ser501

**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:** 6.3

**Predicted Molecular Mass:** 58.4kDa

**Accurate Molecular Mass:** 58kDa 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.

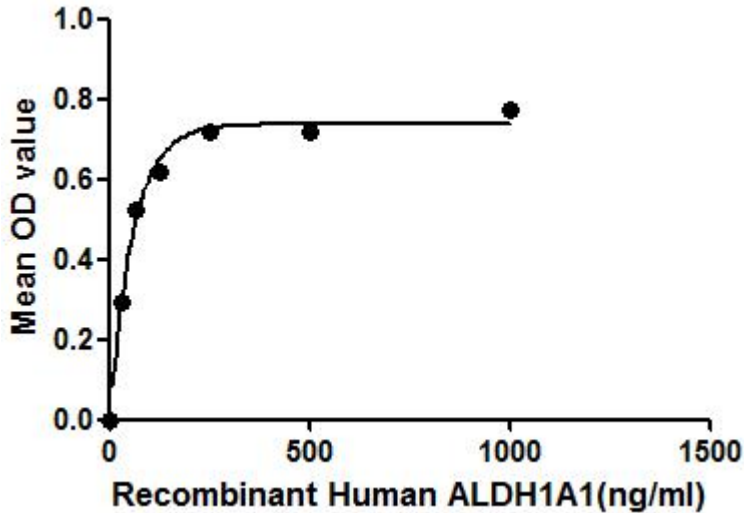
## **[ SEQUENCE ]**

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SSSGTPDLP VLLTDLKIQY TKIFINNEWH DSVSGKKFPV FNPATEEELC
QVEEGDKEDV DKAVKAARQA FQIGSPWRTM DASERGRLLY KLADLIERDR
LLLATMESMN GGKLYSNAYL NDLAGCIKTL RYCAGWADKI QGRTIPIDGN
FFTYTRHEPI GVCQIIPWN FPLVMLIWKI GPALSCGNTV VVKPAEQTPL
TALHVASLIK EAGFPPGVVN IVPGYGPTAG AAISSHMDID KVAFTGSTEV
GKLIKEAAGK SNLKRVTLEL GGKSPCIVLA DADLDNAVEF AHHGVFYHQG
QCCIAASRIF VEESIYDEFV RRSVERAKKY ILGNPLTPGV TQGPQIDKEQ
YDKILDIES GKKEGAKLEC GGGPWGNKGY FVQPTVFSNV TDEMRIAKEE
IFGPVQQIMK FKSLDDVIKR ANNTFYGLSA GVFTKDIDKA ITISSALQAG
TVWVNCYGVV SAQCPFQGGFK MSGNGRELGE YGFHEYTEVK TVTVKISQKN
S
```

## **[ ACTIVITY ]**

ALDH1A1 (Retinal dehydrogenase 1) which belongs to the aldehyde dehydrogenase family, is the next enzyme after alcohol dehydrogenase in the major pathway of alcohol metabolism. ALDH1A1 converts retinaldehyde to retinoic acid. IL21 (interleukin 21) was identified as an interactor of ALDH1A1 through Affinity Capture-MS. Thus a binding ELISA assay was conducted to detect the interaction of recombinant human ALDH1A1 and recombinant human IL21. Briefly, ALDH1A1 were diluted serially in PBS, with 0.01%BSA (pH 7.4). Duplicate samples of 100 ul ALDH1A1 were then transferred to IL21-coated microtiter wells and incubated for 2h at 37°C. Wells were washed with PBST and incubated for 1h with anti-ALDH1A1 pAb, then aspirated and washed 3 times. After incubation with HRP labelled secondary antibody, wells were aspirated and washed 3 times. With the addition of substrate solution, wells were incubated 15-25 minutes at 37°C.

Finally, add 50  $\mu$ L stop solution to the wells and read at 450 nm immediately. The binding activity of ALDH1A1 and IL21 was shown in Figure 1, and this effect was in a dose dependent manner.



**Figure 1. The binding activity of ALDH1A1 with IL21**

The activity of recombinant human ALDH1A1 was also measured by its ability to produce NADH during the oxidation of propionaldehyde. The reaction was performed in 0.1 M Tris, 0.1 M KCl, 2mM DTT, pH 8.5 (assay buffer), initiated by addition 50  $\mu$ L of various concentrations of ALDH1A1 (diluted by assay buffer) to 50  $\mu$ L of substrate mixture 2 mM beta-NAD and 20 mM propionaldehyde. The final well serves as a negative control with no ALDH1A1, replaced with 50  $\mu$ L assay buffer. Read at a wavelength of 340 nm in kinetic mode for 5 minutes. The specific activity of recombinant human ALDH1A1 is > 130 pmol/min/ $\mu$ g.

Specific Activity (pmol/min/ $\mu$ g)=

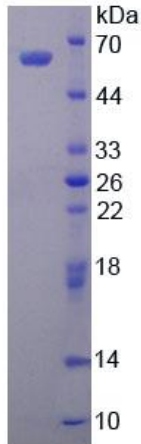
$$\frac{\text{Adjusted } V_{\text{max}}^* (\text{OD}/\text{min}) \times \text{well volume (L)} \times 10^{12} \text{ pmol/mol}}{\text{ext. coeff}^{**} (\text{M}^{-1}\text{cm}^{-1}) \times \text{path corr.}^{***} (\text{cm}) \times \text{amount of enzyme } (\mu\text{g})}$$

\*Adjusted for Substrate Blank

\*\*Using the extinction coefficient 6220  $\text{M}^{-1}\text{cm}^{-1}$

\*\*\*Using the path correction 0.320 cm

**[ IDENTIFICATION ]**



**Figure 2. SDS-PAGE**

**Sample: Active recombinant ALDH1A1, Human**

**[ 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.