APD299Hu01 10μg
Active Cytochrome P450 3A4 (CYP3A4)
Organism Species: *Homo sapiens* (Human)
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: Met1~Ala503
Tags: N-terminal His-tag
Purity: >90%
Endotoxin Level: <1.0EU per 1μg (determined by the LAL method).
Buffer Formulation: PBS, pH7.4, containing 0.01% Sarcosyl, 5%Trehalose .
Original Concentration: 50μg/mL
Applications: Activity Assays.
(May be suitable for use in other assays to be determined by the end user.)
Predicted isoelectric point: 8.2
Predicted Molecular Mass: 61.0kDa
Accurate Molecular Mass: 61kDa as determined by SDS-PAGE reducing conditions.

[USAGE]

Reconstitute in ddH_2O to a concentration of 0.1-0.2 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]

MALIPDLAMETWLLLAVSLVLLYLYGTHSHGLFKKLGIPGPTPLPFLGNILSYHKGFCMFDMECHK
KYGKVWGFYDGQQPVLAITDPDMIKTVLVKECYSVFTNRRPFGPVGFMKSAISIAEDEEWKRLR
SLLSPTFTSGKLKEMVPIIAQYGDVLVRNLRREAETGKPVTLKDVFGAYSMDVITSTSFGVNIDSL
NNPQDPFVENTKKLLRFDFLDPFFLSITVFPFLIPILEVLNICVFPREVTNFLRKSVKRMKESRLEDT
QKHRVDFLQLMIDSQNSKETESHKALSDLELVAQSIIFIFAGYETTSSVLSFIMYELATHPDVQQKL
QEEIDAVLPNKAPPTYDTVLQMEYLDMVVNETLRLFPIAMRLERVCKKDVEINGMFIPKGVVV
MIPSYALHRDPKYWTEPEKFLPERFSKKNKDNIDPYIYTPFGSGPRNCIGMRFALMNMKLALIRV
LQNFSFKPCKETQIPLKLSLGGLLQPEKPVVLKVESRDGTVSGA

[ACTIVITY]

Cytochrome P450 3A4 (CYP3A4), a member of the cytochrome P450 superfamily of heme - containing enzymes, is predominantly expressed in the human liver and small intestine, serving as a key player in xenobiotic metabolism and endobiotic biotransformation. It catalyzes the oxidation of over 50% of clinically used drugs, including statins, immunosuppressants, and anticancer agents, by adding oxygen atoms to hydrophobic substrates, thereby facilitating their excretion. CYP3A4 expression and activity are highly variable among individuals due to genetic polymorphisms, environmental inducers (e.g., rifampicin, phenobarbital) and inhibitors (e.g., ketoconazole, grapefruit juice). Beyond drug metabolism, it participates in the breakdown of endogenous compounds such as steroids and bile acids, maintaining physiological homeostasis. Notably, CYP3A4 and CYP1A1, both phase I metabolic enzymes, exhibit potential functional binding to synergistically modulate the metabolism of shared substrates, with their interaction being a critical part of the hepatic metabolic network.To detect the activity of recombinant CYP3A4, a functional ELISA assay was performed to evaluate the interaction between recombinant human CYP3A4 and recombinant human

CYP1A1.Briefly, biotin-linked CYP3A4 were diluted serially in PBS, with 0.01% BSA (pH 7.4). Duplicate samples of 100µl were then transferred to CYP1A1-coated microtiter wells and incubated for 1h at 37℃. Wells were washed with PBST 3 times and incubation with Streptavidin-HRP for 30min, then wells were aspirated and washed 5 times. With the addition of substrate solution, wells were incubated 15-25 minutes at 37℃. Finally, add 50µl stop solution to the wells and read at 450nm immediately. The binding activity of CYP3A4 and CYP1A1 was shown in Figure 1, the EC50 for this effect is 0.03849µg/mL.

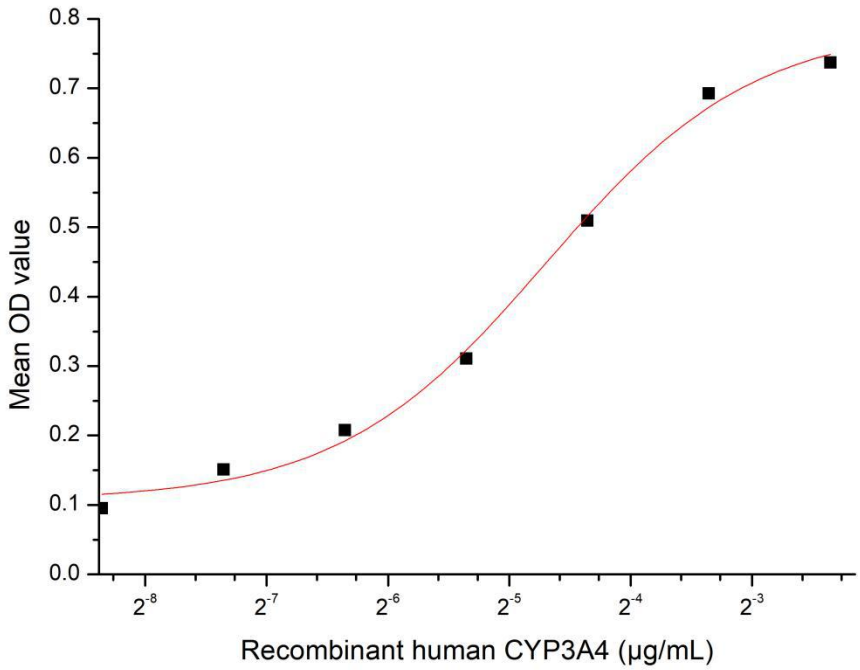

Figure 1. The binding activity of CYP3A4 and CYP1A1

[IDENTIFICATION]

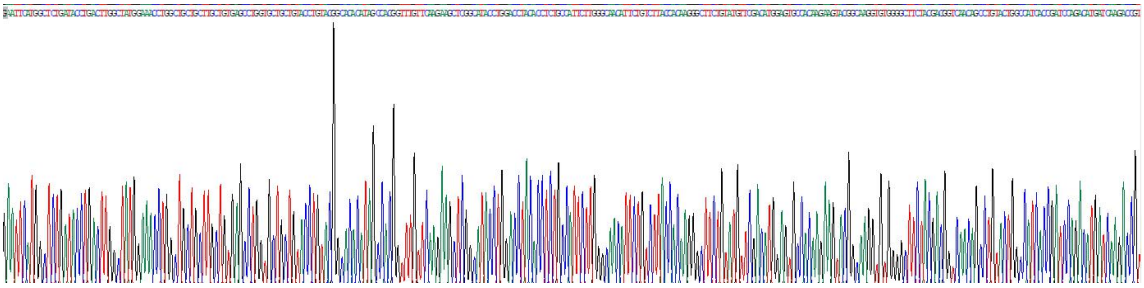

Figure 2. Gene Sequencing (extract)

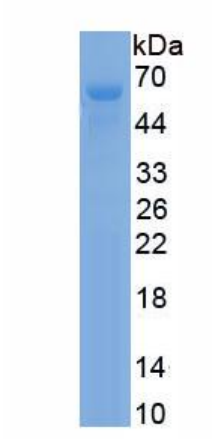

Figure 3. SDS-PAGE

Sample: Active recombinant CYP3A4, 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.