

**APA751Fe61 100µg**  
**Active Programmed Cell Death Protein 1 (PD1)**  
**Organism Species: *Felis catus*; *Feline (Cat)***  
***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:** Eukaryotic expression.

**Host:** 293F cell

**Residues:** Pro31~Thr143

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

**Purity:** >90%

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

**Buffer Formulation:** PBS, pH7.4, containing 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:** 5.6

**Predicted Molecular Mass:** 14.4kDa

**Accurate Molecular Mass:** 22-30kDa as determined by SDS-PAGE reducing conditions.

Phenomenon explanation:

The possible reasons that the actual band size differs from the predicted are as follows:

1. Splice variants: Alternative splicing may create different sized proteins from the same gene.
2. Relative charge: The composition of amino acids may affects the charge of the protein.
3. Post-translational modification: Phosphorylation, glycosylation, methylation etc.
4. Post-translation cleavage: Many proteins are synthesized as pro-proteins, and then cleaved to give the active form.
5. Polymerization of the target protein: Dimerization, multimerization etc.

## **[ 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.

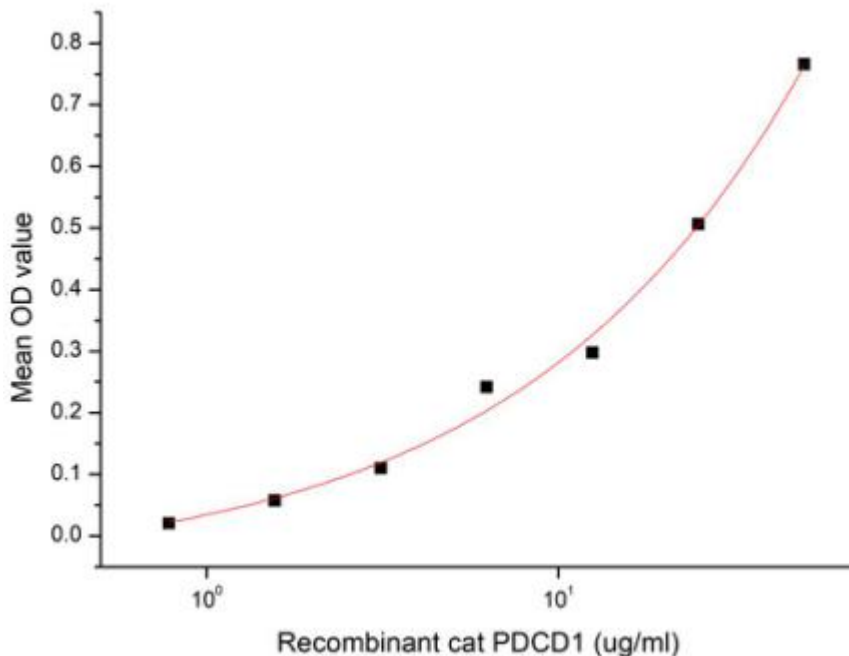
## **[ SEQUENCE ]**

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PWSPLTFSPAQLTVLEGENATFVCHLPDVPEFVLNWWYRVSPRNQTDKLAAFQENHTEPGKDRRFRVTRLPSGQDFHTTILA  
AQLNDSGIYLCGAIYLPNTQIYESPRAELT
```

## **[ ACTIVITY ]**

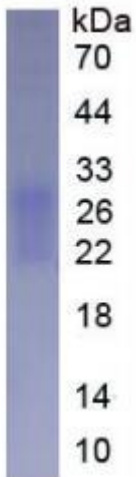
PD-1 (Programmed Death-1 receptor), also known as CD279, is a receptor expressed on T cells responsible for modulating T cell activation. Like CTLA-4, PD-1 is classified as an immune checkpoint inhibitory receptor. When PD-1 protein binds to PD-L1, it initiates a negative signaling cascade inhibiting activation of T cells. The cytoplasmic tail contains two tyrosine residues that form the immunoreceptor tyrosine-based inhibitory motif (ITIM) and immunoreceptor tyrosine-based switch motif (ITSM) that are important for mediating PD-1 signaling. Normally, PD-1 helps keep T cells from attacking other cells in the body. However, many cancers take advantage of this by expressing high amounts of PD-L1 allowing cancer cells to evade the body's own immune response. Blocking the PD-1:PD-L1 interaction has proven successful in treating many different cancer types. A functional binding ELISA assay was conducted to detect the interaction of

recombinant cat PDCD1 and recombinant rat PDCD1LG2. Briefly, biotin-linked PDCD1 were diluted serially in PBS, with 0.01% BSA (pH 7.4). Duplicate samples of 100  $\mu$ l were then transferred to PDCD1LG2-coated microtiter wells and incubated for 1h at 37 °C. 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 °C. Finally, add 50  $\mu$ l stop solution to the wells and read at 450 nm immediately. The binding activity of PDCD1 and PDCD1LG2 was shown in Figure 1, and this effect was in a dose dependent manner.



**Figure 1. The binding activity of recombinant cat PDCD1 and recombinant rat PDCD1LG2**

**[ IDENTIFICATION ]**



**Figure 2. SDS-PAGE**

**Sample: Active recombinant PD1, Cat**

**[ 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.