

**APA196Mu61 100µg**  
**Active Galactosidase Beta (GLb)**  
**Organism Species: *Mus musculus* (Mouse)**  
***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:** Ile25~Ser647

**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:** 7.3

**Predicted Molecular Mass:** 72.2kDa

**Accurate Molecular Mass:** 80kDa 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.

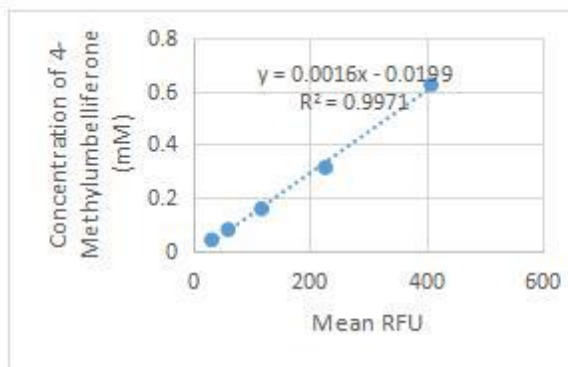
## **[ SEQUENCE ]**

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IS
```

## **[ ACTIVITY ]**

GLB1 is a lysosomal beta -galactosidase that hydrolyzes the terminal beta -galactose from ganglioside and keratan sulfate. Defects in this gene are the causes of lysosomal storage diseases for GM1-gangliosidosis and Morquio B syndrome (also known as mucopolysaccharidosis IVB). In GM1 gangliosidosis, GM1 ganglioside accumulates in the neurons of the central nervous system, because of the deficiency of lysosomal beta -galactosidase activity. GM1 gangliosidosis demonstrates varying degrees of clinical severity but is invariably

fatal, and children with the most common and severe form of GM1 gangliosidosis usually die within 3 years of birth. Morquio B syndrome patients are neurologically normal, but display severe skeletal dysostosis multiplex because of an accumulation of keratan sulfate. The activity assay of GLB1 was measured by its ability to cleave a peptide substrate, 4-Methylumbelliferyl-beta -D-galactopyranoside. The reaction was performed in 50 mM Sodium Citrate, pH 3.5 ( Assay Buffer), initiated by addition 50  $\mu$  L of 1.5 ug/ml uPA (diluted by Assay Buffer) to 50  $\mu$ L of 1.2 mM Substrate. Read at excitation and emission wavelengths of 365 nm and 445 nm (top read), respectively, in kinetic mode for 5 minutes. The specific activity of recombinant mouse GLB1 is >17000 pmol/min/ $\mu$ g.



RFU	4-Methylumbelliferone (mM)
407.3912	0.625
226.2912	0.3125
117.1912	0.15625
59.9312	0.078125
31.6012	0.0390625

**Figure 1. The standard curve of 4-Methylumbelliferone**

One unit of enzyme activity is defined as the 1  $\mu$ g of enzyme required to convert 1 pmol of 4-Methylumbelliferyl-beta -D-galactopyranoside to 4-Methylumbelliferone in 1 min.

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

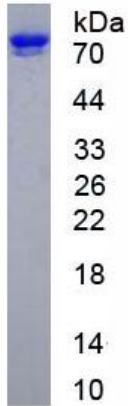
$\Delta$ RFU=Adjusted for Substrate Blank

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

T= Time

N=Amount of enzyme

**[ IDENTIFICATION ]**



**Figure 2. SDS-PAGE**

**Sample: Active recombinant GLb, Mouse**

**[ 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.