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APE112Mu61 100µg Active Ephrin B2 (EFNB2) Organism Species: *Mus musculus (Mouse) Instruction manual*

FOR RESEARCH USE ONLY NOT FOR USE IN CLINICAL DIAGNOSTIC PROCEDURES

13th Edition (Revised in Aug, 2023)

[PROPERTIES]

Source: Eukaryotic expression.

Host: 293F cell

Residues: Arg29~Glu227

Tags: N-terminal His Tag and C-terminal Fc Region of Human IgG1

Purity: >95%

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: 7.9

Predicted Molecular Mass: 48.8kDa

Accurate Molecular Mass: 60kDa 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.

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[<u>USAGE</u>]

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]

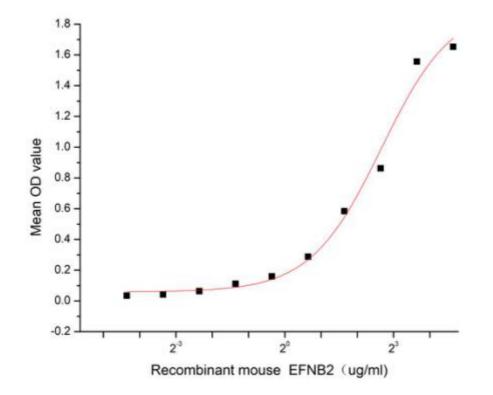
RS IVLEPIYWNS SNSKFLPGQG LVLYPQIGDK LDIICPKVDS KTVGQYEYYK VYMVDKDQAD RCTIKKENTP LLNCARPDQD VKFTIKFQEF SPNLWGLEFQ KNKDYYIIST SNGSLEGLDN QEGGVCQTRA MKILMKVGQD ASSAGSARNH GPTRRPELEA GTNGRSSTTS PFVKPNPGSS TDGNSAGHSG NNLLGSE

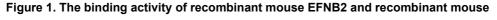
[<u>ACTIVITY</u>]

EphrinB2 (EFNB2), a 40 kDa member of the Ephrin-B family, is a pivotal bidirectional signaling molecule ubiquitously expressed in mammals and is crucial in angiogenesis during development and disease progression. Besides, EFNB2 is expressed at abnormally high levels in some neoplasms, such as squamous cell carcinoma of the head and neck and colorectal cancer. Its overexpression is associated with the malignant progression of tumors. Ephrin Type B Receptor 3 (EPHB3) has been identified as an interactor of EFNB2, thus a functional binding ELISA assay was conducted to detect the interaction of recombinant mouse EFNB2 and recombinant mouse EPHB3. Briefly, biotin-linked EFNB2 were diluted serially in PBS, with 0.01% BSA (pH 7.4). Duplicate samples of 100 ul were then

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transferred to EPHB3-coated microtiter wells and incubated for 1h at 37 $^\circ$ 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 $^\circ$ C. Finally, add 50 μ l stop solution to the wells and read at 450 nm immediately. The binding activity of recombinant mouse EFNB2 and recombinant mouse EPHB3 was shown in Figure 1, the EC50 for this effect is 6.3 ug/mL.





EPHB3

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[IDENTIFICATION]

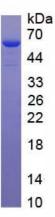


Figure 2. SDS-PAGE

Sample: Active recombinant EFNB2, 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.