

Active Nesfatin 1 (NES1) Instruction Manual

SBPA106Mu01

Mus musculus (Mouse)

Buffer Formulation

20mM Tris, 150mM NaCl, pH8.0, containing 1mM EDTA, 1mM DTT, 0.01% SKL, 5% Trehalose and Proclin300.

Traits

Freeze-dried powder

Purity

> 97%

Isoelectric Point

5.8

Applications

Cell culture; Activity Assays.

ACTIVITY TEST

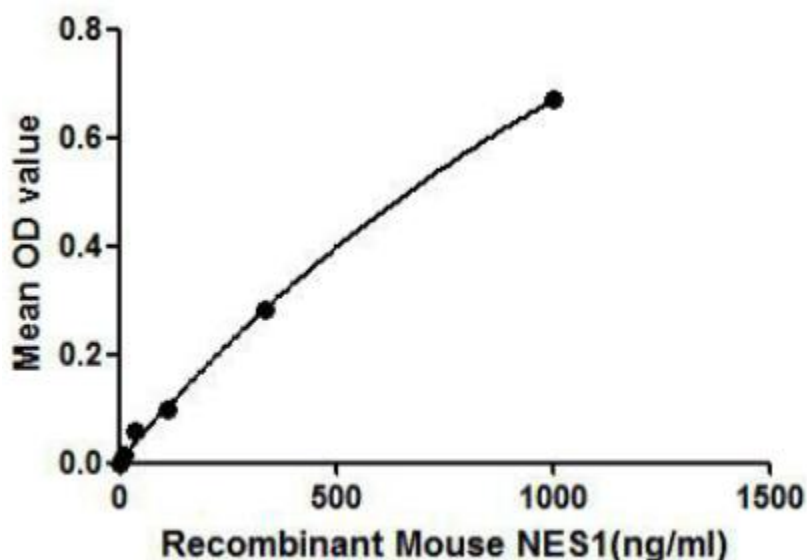


Figure 1. The binding activity of NES1 with GADD45A.

Nucb2 (Nucleobindin-2), a calcium-binding protein, is further cleaved into NES1 (Nesfatin-1). NES1 is an anorexigenic peptide and seems to participate in hypothalamic pathways regulating food intake and energy homeostasis, acting in a leptin-independent manner. GADD45A (Growth arrest and DNA damage-inducible protein GADD45 alpha) has been identified as an interactor of Nucb2. Besides, rat GADD45A shares similarities with mouse NES1 in amino acid sequence with the identity of 98%. Thus a binding ELISA assay was conducted to detect the interaction of recombinant mouse NES1 and recombinant rat GADD45A. Briefly, NES1 were diluted serially in PBS, with 0.01%BSA (pH 7.4). Duplicate samples of 100uL NES1 were then transferred to GADD45A-coated

microtiter wells and incubated for 2h at 37°C. Wells were washed with PBST and incubated for 1h with anti-NES1 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 50µL stop solution to the wells and read at 450nm immediately. The binding activity of NES1 and GADD45A was shown in Figure 1, and this effect was in a dose dependent manner.

USAGE

Reconstitute in 20mM Tris, 150mM NaCl (pH8.0) to a concentration of 0.1-1.0 mg/mL. Do not vortex.

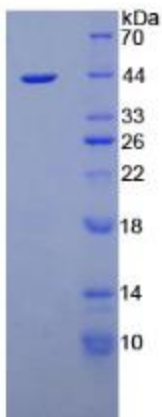
STORAGE

Avoid repeated freeze/thaw cycles. Store at 2-8°C for one month. Aliquot and store at -80°C for 12 months.

STABILITY

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.

Image



SDS-PAGE Image

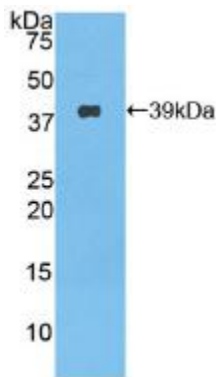


Figure. Western Blot; Sample: Recombinant NES1, Mouse.

[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.