

Active Ribonuclease P (RNASEP) Instruction Manual

SBPA098Ra01

Rattus norvegicus (Rat)

Buffer Formulation	PBS, pH7.4, containing 0.01% SKL, 5% Trehalose.
Traits	Freeze-dried powder
Purity	> 97%
Isoelectric Point	6.2
Applications	Cell culture; Activity Assays.

ACTIVITY TEST

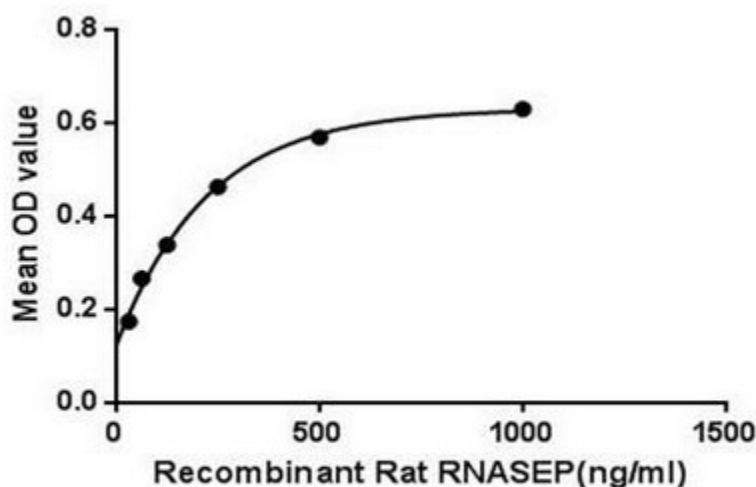


Figure 1. The binding activity of RNASEP with NPM.

Ribonuclease P is a site specific endonuclease that generates mature tRNAs by catalysing the removal of the 5'-leader sequence from pre-tRNA to produce the mature 5'-terminus. It can also cleave other RNA substrates such as 4.5S RNA. In bacteria, RNase P consists of two components: a large RNA (about 400 base pairs) encoded by *rnpB*, and a small protein (119 to 133 amino acids) encoded by *rnpA*. The RNA moiety of RNase P carries the catalytic activity; the protein component plays an auxiliary, but essential, role in vivo by binding to the 5'-leader sequence and broadening the substrate specificity of the ribozyme. The sequence of *rnpA* is not highly conserved, however there is, in the central part of the protein, a conserved basic region. Besides, Nucleophosmin (NPM) has been identified as an interactor of RNASEP, thus a binding ELISA assay was conducted to detect the interaction of recombinant rat RNASEP and recombinant rat NPM. Briefly,

RNASEP were diluted serially in PBS, with 0.01% BSA (pH 7.4). Duplicate samples of 100uL were then transferred to NPM-coated microtiter wells and incubated for 2h at 37°C. Wells were washed with PBST and incubated for 1h with anti-RNASEP 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 RNASEP and NPM was shown in Figure 1, and this effect was in a dose dependent manner.

USAGE

Reconstitute in 10mM PBS (pH7.4) to a concentration of 0.1-1.0 mg/mL. Do not vortex.

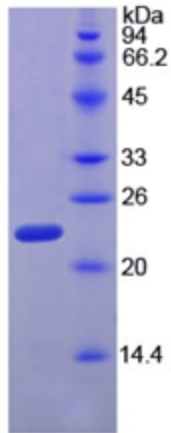
STORAGE

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

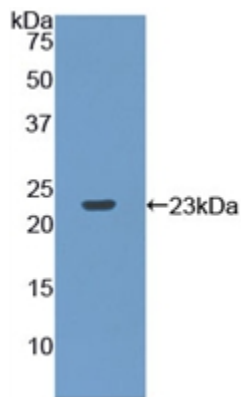


Figure. Western Blot; Sample: Recombinant RNASEP, Rat.

[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.