**APA198Ra01 100μg**
**Active Ribonuclease P (RNASEP)**
**Organism Species: Rattus norvegicus (Rat)**
***Instruction manual***

FOR IN VITRO USE AND RESEARCH USE ONLY
NOT FOR USE IN CLINICAL DIAGNOSTIC PROCEDURES

1st Edition (Apr, 2016)

## [ PROPERTIES ]

**Source:** Prokaryotic expression.
**Host:** *E. coli*
**Residues:** Met91~Asp259
**Tags:** N-terminal His-tag
**Purity:** >98%
**Buffer Formulation:** 20mM Tris, 150mM NaCl, pH8.0, containing 0.05% sarcosyl and 5% trehalose.
**Applications:** Cell culture; Activity Assays.
(May be suitable for use in other assays to be determined by the end user.)
**Predicted isoelectric point:** 6.2
**Predicted Molecular Mass:** 23.0kDa
**Accurate Molecular Mass:** 23kDa as determined by SDS-PAGE reducing conditions.

## [ USAGE ]

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

## [ STORAGE AND STABILITY ]

**Storage:** Avoid repeated freeze/thaw cycles.
Store at 2-8ºC for one month.
Aliquot and store at -80ºC 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 37ºC 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 ]

MDLSLNLDSK
KYRRISWSFK EKKPLKFDFL LAWHHTGTEE STMMSYFSKY QIREHQPKVA
LSTVRDLQCP VLQSSSLAGE PEEACNALEF FDWLGAVFCN ADLNNEPHNF
ISTYCCPQPN TVAAQACLCT ITGFVLPEKI LVLLEQLCHY FDEPKLAPWV
TLTVQGFAD

## [ ACTIVITY ]

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 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℃. 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℃. Finally, add 50μL stop solution to the wells and read at 450nm immediately. The binding activity of of RNASEP and NPM was shown in Figure 1, and this effect was in a dose dependent manner.

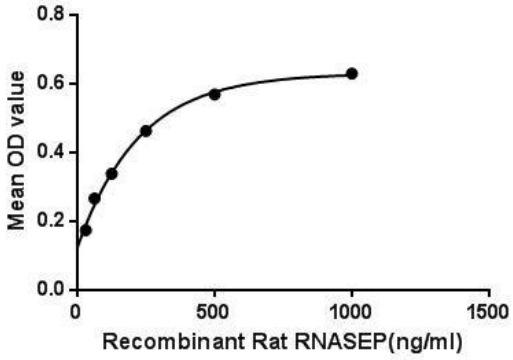


**Figure 1. The binding activity of RNASEP with NPM.**

## [ IDENTIFICATION ]

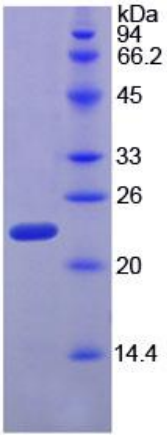


**Figure 2. SDS-PAGE**

**Sample: Active recombinant RNASEP, Rat**

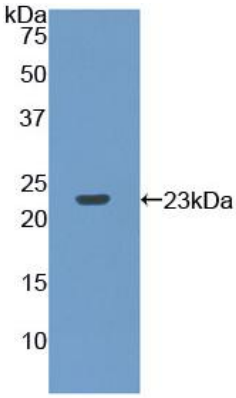


**Figure 3. Western Blot**

**Sample: Recombinant RNASEP, Rat;**

**Antibody: Rabbit Anti-Rat RNASEP Ab (PAA198Ra01)**