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APA837Mu01 100µg Active Nitric Oxide Synthase 2, Inducible (NOS2) Organism Species: *Mus musculus (Mouse) Instruction manual*

FOR RESEARCH USE ONLY NOT FOR USE IN CLINICAL DIAGNOSTIC PROCEDURES

1st Edition (Apr, 2016)

[PROPERTIES]

Source: Prokaryotic expression. Host: *E. coli* Residues: Asp43~Thr213 Tags: N-terminal His-tag Purity: >98% Endotoxin Level: <1.0EU per 1µg (determined by the LAL method). 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: 7.8 Predicted Molecular Mass: 20.6kDa

Accurate Molecular Mass: 20kDa as determined by SDS-PAGE reducing conditions.

[<u>USAGE</u>]

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 2-8°C for one month.

Aliquot and store at -80°C for 12 months.

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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.

[<u>SEQUENCE</u>]

DPKSHQNG SPQLLTGTAQ NVPESLDKLH VTSTRPQYVR IKNWGSGEIL HDTLHHKATS DFTCKSKSCL GSIMNPKSLT RGPRDKPTPL EELLPHAIEF INQYYGSFKE AKIEEHLARL EAVTKEIETT GTYQLTLDEL IFATKMAWRN APRCIGRIQW SNLQVFDARN CST

[<u>ACTIVITY</u>]

Nitric oxide synthase 2, inducible (NOS2) is a member of Nitric oxide synthases (NOSs) family. Nitric oxide synthases (NOSs) are a family of enzymes catalyzing the production of nitric oxide (NO) from L-arginine. NO is an important cellular signaling molecule. It helps modulate vascular tone, insulin secretion, airway tone, and peristalsis, and is involved in angiogenesis and neural development. Besides, Ubiquitin Carboxyl Terminal Hydrolase L5 (UCHL5) has been identified as an interactor of NOS2, thus a binding ELISA assay was conducted to detect the interaction of recombinant mouse NOS2 and recombinant mouse UCHL5. Briefly, NOS2 was diluted serially in PBS with 0.01% BSA (pH 7.4). Duplicate samples of 100uL were then transferred to UCHL5-coated microtiter wells and incubated for 2h at 37 °C . Wells were washed with PBST and incubated for 1h with anti-NOS2 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 NOS2 and UCHL5 was shown in Figure 1, and this effect was in a dose dependent manner.

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Figure 1. The binding activity of NOS2 with UCHL5.





Figure 2. SDS-PAGE

Sample: Active recombinant NOS2, Mouse

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Figure 3. Western Blot Sample: Recombinant NOS2, Mouse; Antibody: Rabbit Anti-Mouse NOS2 Ab (PAA837Mu01)

[IMPORTANT NOTE]

The kit is designed for in vitro and research use only, we will not be responsible for any issue if the kit was used in clinical diagnostic or any other procedures.