

DESCRIPTION

Source Mouse myeloma cell line, NS0-derived
Glu38-Leu278, with a C-terminal 6-His tag
Accession # NP_006577

N-terminal Sequence Analysis Glu38

Predicted Molecular Mass 26.9 kDa

SPECIFICATIONS

SDS-PAGE 37-40 kDa, reducing conditions

Activity Measured by its binding ability in a functional ELISA.
Immobilized Recombinant Human PRAT4A at 2 µg/mL can bind Recombinant Human TLR4/MD-2 Complex (Catalog # 3146-TM) with an apparent $K_D < 15$ nM.

Endotoxin Level <0.01 EU per 1 µg of the protein by the LAL method.

Purity >95%, by SDS-PAGE under reducing conditions and visualized by silver stain.

Formulation Lyophilized from a 0.2 µm filtered solution in PBS. See Certificate of Analysis for details.

PREPARATION AND STORAGE

Reconstitution Reconstitute at 300 µg/mL in PBS.

Shipping The product is shipped at ambient temperature. Upon receipt, store it immediately at the temperature recommended below.

Stability & Storage Use a manual defrost freezer and avoid repeated freeze-thaw cycles.

- 12 months from date of receipt, -20 to -70 °C as supplied.
- 1 month, 2 to 8 °C under sterile conditions after reconstitution.
- 3 months, -20 to -70 °C under sterile conditions after reconstitution.

BACKGROUND

PRAT4A (PProtein Associated with Toll-like receptor 4A), also called CNPY3 (Canopy homolog 3) or TNRC5 (trinucleotide repeat-containing 5) is a widely expressed 40 kDa protein that is an intracellular chaperone for Toll-like receptors (TLRs) (1-3). Human PRAT4A cDNA encodes 278 amino acids (aa), including an N-terminal hydrophobic sequence of 26-37 aa (putative signal sequence) and a 250-241 aa mature region (3). A potential 153 aa form has an alternate start site at aa 126. Human PRAT4A shares 90%, 90%, 94%, 95% and 96% aa sequence identity with mouse, rat, bovine, canine and porcine PRAT4A, respectively. A related protein, PRAT4B, shares approximately 40% aa sequence identity, is co-expressed, and is reported to bind TLR4 only if it lacks mature glycosylated structures (4). PRAT4A resides in the endoplasmic reticulum (ER) and is a co-chaperone that provides substrate-specificity to the chaperone gp96, an HSP90 paralog required for proper folding of TLRs (1-3, 5, 6). It binds TLR4, enhances TLR4 N-linked glycosylation, and forms a heterotrimer with TLR4 and its co-receptor, MD2 (6-8). PRAT4A is required for transfer of TLR4 from the ER to the plasma membrane where it recognizes its extracellular ligand, bacterial lipopolysaccharide (LPS) (1-3, 6, 7). PRAT4A is also essential for maturation and trafficking of TLR9 from the ER to endolysosomes in response to its intracellular ligand, unmethylated DNA (1-3, 5-7, 9). PRAT4A deletion, knockdown, or specific mutation in mice abolishes or lowers surface expression of TLR4/MD2, TLR2, TLR1 and RP105/CD180, and abolishes production of RANTES in response to a TLR7 ligand (3, 6-8). PRAT4A enhances Th1 responses and production of inflammatory cytokines in response to TLR ligands, and thus contributes to endotoxic shock (2, 6-8).

References:

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