10320 Camino Santa Fe, Suite G
San Diego, CA 92121
Tel: 858.875.1900 Fax: 858.875.1999

NEDD4 Polyclonal Antibody

Purified Rabbit Polyclonal Antibody (Pab)
Catalog # AP58787

Product Information

Application	WB, IHC-P, IHC-F, IF, ICC, E
Primary Accession	P46934
Reactivity	Rat, Pig, Dog, Bovine
Host	Rabbit
Clonality	Polyclonal
Calculated MW	149114
Physical State	Liquid
Immunogen	KLH conjugated synthetic peptide derived from human SRD5A2
Epitope Specificity	801-900/1319
Isotype	IgG
Purity	affinity purified by Protein A
Buffer	0.01M TBS (pH7.4) with 1% BSA, 0.02% Proclin300 and 50% Glycerol.
SUBCELLULAR LOCATION	Cytoplasm. Cell membrane; Peripheral membrane protein. Note=Recruited to the plasma membrane by GRB10. Once complexed with GRB10 and IGF1R, follows IGF1R internalization, remaining associated with early endosomes. Uncouples from IGF1R-containing endosomes before the sorting of the receptor to the lysosomal compartment. May be recruited to exosomes by NDFIP1..
SIMILARITY	Contains 1 HECT (E6AP-type E3 ubiquitin-protein ligase) domain.Contains 4 WW domains.
SUBUNIT	Interacts with UBE2D2. Binds SCNN1A, SCNN1B and SCNN1G. Binds, in vitro, through the WW2 and WW3 domains, to neural isoforms of ENAH that contain the PPSY motif. Interacts with BEAN1, LITAF, RNF11, WBP1, WBP2, TMEPAI and PRRG2 (By similarity). Interacts with NDFIP1 and NDFIP2; this interaction activates the E3 ubiquitin-protein ligase and may induce its recruitment to exosomes. Interaction with PTEN is questionable according to PubMed:18562292. Interacts with viral proteins that contain a late- budding motif P-P-P-Y. This interaction is essential for viral particle budding of a lot of retroviruses, like HTLV-1 Gag and MLV Gag. Interacts (via C2 domain) with GRB10 (via SH2 domain). Interacts with ERBB4. Interacts with TNIK; the interaction is direct, allows the TNIK-dependent recruitment of RAP2A and its ubiquitination by NEDD4. Interacts (via WW3 domain) with TNK2; EGF promotes this interaction. Interacts (via WW3 domain) with FGFR1 (via C-terminus).
Post-translational modifications	Auto-ubiquitinated.
Important Note	This product as supplied is intended for research use only, not for use in human, therapeutic or diagnostic applications.
Background Descriptions	E3 ubiquitin-protein ligase which accepts ubiquitin from an E2 ubiquitin-conjugating enzyme in the form of a thioester and then directly transfers the ubiquitin to targeted substrates. Involved in the pathway leading to the degradation of VEGFR-2/KDFR, independently of its ubiquitin-ligase activity. Monoubiquitinates IGF1R at multiple sites, thus leading to receptor

internalization and degradation in lysosomes. According to PubMed:18562292 the direct link between NEDD4 and PTEN regulation through polyubiquitination described in PubMed:17218260 is questionable. Involved in ubiquitination of ERBB4 intracellular domain E4ICD. Involved in the budding of many viruses. Part of a signaling complex composed of NEDD4, RAP2A and TNIK which regulates neuronal dendrite extension and arborization during development. Ubiquitinates TNK2 and regulates EGF-induced degradation of EGFR and TNF2.

Additional Information

Gene ID	4734
Other Names	E3 ubiquitin-protein ligase NEDD4, 2.3.2.26, Cell proliferation-inducing gene 53 protein, HECT-type E3 ubiquitin transferase NEDD4, Neural precursor cell expressed developmentally down-regulated protein 4, NEDD-4, NEDD4, KIAA0093, NEDD4-1
Dilution	WB=1:500-2000,IHC-P=1:100-500,IHC-F=1:100-500,ICC=1:100-500,IF=1:100-500,Flow-Cyt=3ug/test,ELISA=1:5000-10000
Format	0.01M TBS(pH7.4) with 1% BSA, 0.09% (W/V) sodium azide and 50% Glyce
Storage	Store at -20 °C for one year. Avoid repeated freeze/thaw cycles. When reconstituted in sterile pH 7.4 0.01M PBS or diluent of antibody the antibody is stable for at least two weeks at 2-4 °C.

Protein Information

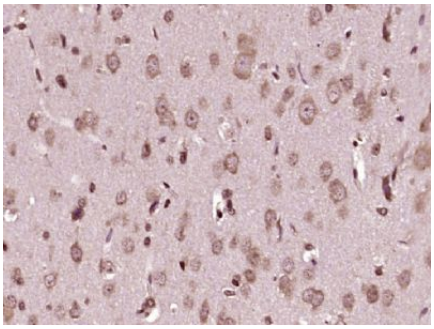
Name	NEDD4
Synonyms	KIAA0093, NEDD4-1, RPF1 {ECO:0000303|Pub
Function	E3 ubiquitin-protein ligase which accepts ubiquitin from an E2 ubiquitin-conjugating enzyme in the form of a thioester and then directly transfers the ubiquitin to targeted substrates. Specifically ubiquitinates 'Lys-63' in target proteins (PubMed:19920177, PubMed:21399620, PubMed:23644597). Involved in the pathway leading to the degradation of VEGFR-2/KDFR, independently of its ubiquitin-ligase activity. Monoubiquitinates IGF1R at multiple sites, thus leading to receptor internalization and degradation in lysosomes (By similarity). Ubiquitinates FGFR1, leading to receptor internalization and degradation in lysosomes (PubMed:21765395). Promotes ubiquitination of RAPGEF2 (PubMed:11598133). According to PubMed:18562292 the direct link between NEDD4 and PTEN regulation through polyubiquitination described in PubMed:17218260 is questionable. Involved in ubiquitination of ERBB4 intracellular domain E4ICD (By similarity). Part of a signaling complex composed of NEDD4, RAP2A and TNIK which regulates neuronal dendrite extension and arborization during development (By similarity). Ubiquitinates TNK2 and regulates EGF-induced degradation of EGFR and TNF2 (PubMed:20086093). Ubiquitinates BRAT1 and this ubiquitination is enhanced in the presence of NDFIP1 (PubMed:25631046). Ubiquitinates DAZAP2, leading to its proteasomal degradation (PubMed:11342538). Ubiquitinates POLR2A (PubMed:19920177). Functions as a platform to recruit USP13 to form an NEDD4-USP13 deubiquitination complex that plays a critical role in cleaving the 'Lys-48'-linked ubiquitin chains of VPS34 and then stabilizing

VPS34, thus promoting the formation of autophagosomes (PubMed:32101753).

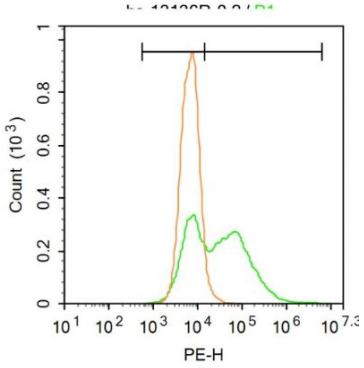
Cellular Location

Cytoplasm. Nucleus. Cell membrane {ECO:0000250|UniProtKB:P46935}; Peripheral membrane protein {ECO:0000250|UniProtKB:P46935}. Note=Predominantly cytoplasmic but also located in the nucleus (PubMed:11342538). Recruited to the plasma membrane by GRB10. Once complexed with GRB10 and IGF1R, follows IGF1R internalization, remaining associated with early endosomes. Uncouples from IGF1R-containing endosomes before the sorting of the receptor to the lysosomal compartment (By similarity). May be recruited to exosomes by NDFIP1 (PubMed:18819914). {ECO:0000250|UniProtKB:P46935, ECO:0000269|PubMed:11342538, ECO:0000269|PubMed:18819914}

Images



Paraformaldehyde-fixed, paraffin embedded (rat brain tissue); Antigen retrieval by boiling in sodium citrate buffer (pH6.0) for 15min; Block endogenous peroxidase by 3% hydrogen peroxide for 20 minutes; Blocking buffer (normal goat serum) at 37°C for 30min; Antibody incubation with (NEDD4) Polyclonal Antibody, Unconjugated (AP58787) at 1:400 overnight at 4°C, followed by operating according to SP Kit(Rabbit) (sp-0023) instructionsand DAB staining.



Blank control:A549.
Primary Antibody (green line): Rabbit Anti-NEDD4 antibody (AP58787)
Dilution: 1 µg /10^6 cells;
Isotype Control Antibody (orange line): Rabbit IgG .
Secondary Antibody : Goat anti-rabbit IgG-PE
Dilution: 3 µg /test.
Protocol
The cells were fixed with 4% PFA (10min at room temperature)and then permeabilized with 20% PBST for 20 min at room temperature. The cells were then incubated in 5% BSA to block non-specific protein-protein interactions for 30 min at at room temperature .Cells stained with Primary Antibody for 30 min at room temperature. The secondary antibody used for 40 min at room temperature. Acquisition of 20,000 events was performed.