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Anti-mTOR (Phospho-S2448) Antibody

Catalog # AP60683

Product Information

Application	WB, IHC, IF/IC
Primary Accession	P42345
Reactivity	Human
Host	Rabbit
Clonality	Polyclonal
Calculated MW	288892

Additional Information

Gene ID	2475
Other Names	FRAP; FRAP1; FRAP2; RAFT1; RAPT1; Serine/threonine-protein kinase mTOR; FK506-binding protein 12-rapamycin complex-associated protein 1; FKBP12-rapamycin complex-associated protein; Mammalian target of rapamycin; mTOR; Mechanistic target of rapamycin; Rapamycin and FKBP12 target 1; Rapamycin target protein 1
Target/Specificity	KLH-conjugated synthetic phosphopeptide corresponding to residues surrounding S2448 of human mTOR protein. The exact sequence is proprietary.
Dilution	WB~~WB (1/500 - 1/1000), IHC (1/100 - 1/200), IF/IC (1/100 - 1/500), IP (1/10 - 1/100) IHC~~WB (1/500 - 1/1000), IHC (1/100 - 1/200), IF/IC (1/100 - 1/500), IP (1/10 - 1/100) IF/IC~~N/A
Format	Liquid in 0.42% Potassium phosphate, 0.87% Sodium chloride, pH 7.3, 30% glycerol, and 0.01% sodium azide.
Storage	Store at -20 °C.Stable for 12 months from date of receipt

Protein Information

Name	MTOR (HGNC:3942)
Function	Serine/threonine protein kinase which is a central regulator of cellular metabolism, growth and survival in response to hormones, growth factors, nutrients, energy and stress signals (PubMed:12087098, PubMed:12150925, PubMed:12150926, PubMed:12231510, PubMed:12718876, PubMed:14651849, PubMed:15268862, PubMed:15467718, PubMed:15545625, PubMed:15718470, PubMed:18497260, PubMed:18762023, PubMed:18925875, PubMed:20516213, PubMed:20537536, PubMed:21659604, PubMed:23429703, PubMed:23429704, PubMed:25799227, PubMed:26018084,

PubMed:29150432, PubMed:29236692, PubMed:31112131, PubMed:31601708, PubMed:32561715, PubMed:34519269, PubMed:37751742). MTOR directly or indirectly regulates the phosphorylation of at least 800 proteins (PubMed:15268862, PubMed:15467718, PubMed:17517883, PubMed:18372248, PubMed:18497260, PubMed:18925875, PubMed:20516213, PubMed:21576368, PubMed:21659604, PubMed:23429704, PubMed:30171069, PubMed:29236692, PubMed:37751742). Functions as part of 2 structurally and functionally distinct signaling complexes mTORC1 and mTORC2 (mTOR complex 1 and 2) (PubMed:15268862, PubMed:15467718, PubMed:18497260, PubMed:18925875, PubMed:20516213, PubMed:21576368, PubMed:21659604, PubMed:23429704, PubMed:29424687, PubMed:29567957, PubMed:35926713). In response to nutrients, growth factors or amino acids, mTORC1 is recruited to the lysosome membrane and promotes protein, lipid and nucleotide synthesis by phosphorylating key regulators of mRNA translation and ribosome synthesis (PubMed:12087098, PubMed:12150925, PubMed:12150926, PubMed:12231510, PubMed:12718876, PubMed:14651849, PubMed:15268862, PubMed:15467718, PubMed:15545625, PubMed:15718470, PubMed:18497260, PubMed:18762023, PubMed:18925875, PubMed:20516213, PubMed:20537536, PubMed:21659604, PubMed:23429703, PubMed:23429704, PubMed:25799227, PubMed:26018084, PubMed:29150432, PubMed:29236692, PubMed:31112131, PubMed:34519269). This includes phosphorylation of EIF4EBP1 and release of its inhibition toward the elongation initiation factor 4E (eiF4E) (PubMed:24403073, PubMed:29236692). Moreover, phosphorylates and activates RPS6KB1 and RPS6KB2 that promote protein synthesis by modulating the activity of their downstream targets including ribosomal protein S6, eukaryotic translation initiation factor EIF4B, and the inhibitor of translation initiation PDCD4 (PubMed:12087098, PubMed:12150925, PubMed:18925875, PubMed:29150432, PubMed:29236692). Stimulates the pyrimidine biosynthesis pathway, both by acute regulation through RPS6KB1-mediated phosphorylation of the biosynthetic enzyme CAD, and delayed regulation, through transcriptional enhancement of the pentose phosphate pathway which produces 5-phosphoribosyl-1- pyrophosphate (PRPP), an allosteric activator of CAD at a later step in synthesis, this function is dependent on the mTORC1 complex (PubMed:23429703, PubMed:23429704). Regulates ribosome synthesis by activating RNA polymerase III-dependent transcription through phosphorylation and inhibition of MAF1 an RNA polymerase III-repressor (PubMed:20516213). Activates dormant ribosomes by mediating phosphorylation of SERBP1, leading to SERBP1 inactivation and reactivation of translation (PubMed:36691768). In parallel to protein synthesis, also regulates lipid synthesis through SREBF1/SREBP1 and LPIN1 (PubMed:23426360). To maintain energy homeostasis mTORC1 may also regulate mitochondrial biogenesis through regulation of PPARGC1A (By similarity). In the same time, mTORC1 inhibits catabolic pathways: negatively regulates autophagy through phosphorylation of ULK1 (PubMed:32561715). Under nutrient sufficiency, phosphorylates ULK1 at 'Ser-758', disrupting the interaction with AMPK and preventing activation of ULK1 (PubMed:32561715). Also prevents autophagy through phosphorylation of the autophagy inhibitor DAP (PubMed:20537536). Also prevents autophagy by phosphorylating RUBCNL/Pacer under nutrient-rich conditions (PubMed:30704899). Prevents autophagy by mediating phosphorylation of AMBRA1, thereby inhibiting AMBRA1 ability to mediate ubiquitination of ULK1 and interaction between AMBRA1 and PPP2CA (PubMed:23524951, PubMed:25438055). mTORC1 exerts a feedback control on upstream growth factor signaling that includes phosphorylation and activation of GRB10 a INSR-dependent signaling suppressor (PubMed:21659604). Among other potential targets mTORC1 may phosphorylate CLIP1 and regulate microtubules (PubMed:12231510). The

mTORC1 complex is inhibited in response to starvation and amino acid depletion (PubMed:12150925, PubMed:12150926, PubMed:24403073, PubMed:31695197). The non-canonical mTORC1 complex, which acts independently of RHEB, specifically mediates phosphorylation of MiT/TFE factors MITF, TFEB and TFE3 in the presence of nutrients, promoting their cytosolic retention and inactivation (PubMed:22343943, PubMed:22576015, PubMed:22692423, PubMed:24448649, PubMed:32612235, PubMed:36608670, PubMed:36697823). Upon starvation or lysosomal stress, inhibition of mTORC1 induces dephosphorylation and nuclear translocation of TFEB and TFE3, promoting their transcription factor activity (PubMed:22343943, PubMed:22576015, PubMed:22692423, PubMed:24448649, PubMed:32612235, PubMed:36608670). The mTORC1 complex regulates pyroptosis in macrophages by promoting GSDMD oligomerization (PubMed:34289345). MTOR phosphorylates RPTOR which in turn inhibits mTORC1 (By similarity). As part of the mTORC2 complex, MTOR transduces signals from growth factors to pathways involved in proliferation, cytoskeletal organization, lipogenesis and anabolic output (PubMed:15268862, PubMed:15467718, PubMed:24670654, PubMed:29424687, PubMed:29567957, PubMed:35926713). In response to growth factors, mTORC2 phosphorylates and activates AGC protein kinase family members, including AKT (AKT1, AKT2 and AKT3), PKC (PRKCA, PRKCB and PRKCE) and SGK1 (PubMed:15268862, PubMed:15467718, PubMed:21376236, PubMed:24670654, PubMed:29424687, PubMed:29567957, PubMed:35926713). In contrast to mTORC1, mTORC2 is nutrient-insensitive (PubMed:15467718). mTORC2 plays a critical role in AKT1 activation by mediating phosphorylation of different sites depending on the context, such as 'Thr-450', 'Ser-473', 'Ser-477' or 'Thr-479', facilitating the phosphorylation of the activation loop of AKT1 on 'Thr-308' by PDPK1/PDK1 which is a prerequisite for full activation (PubMed:15718470, PubMed:21376236, PubMed:24670654, PubMed:29424687, PubMed:29567957). mTORC2 also regulates the phosphorylation of SGK1 at 'Ser-422' (PubMed:18925875). mTORC2 may regulate the actin cytoskeleton, through phosphorylation of PRKCA, PXN and activation of the Rho-type guanine nucleotide exchange factors RHOA and RAC1A or RAC1B (PubMed:15268862). The mTORC2 complex also phosphorylates various proteins involved in insulin signaling, such as FBXW8 and IGF2BP1 (By similarity). May also regulate insulin signaling by acting as a tyrosine protein kinase that catalyzes phosphorylation of IGF1R and INSR; additional evidence are however required to confirm this result in vivo (PubMed:26584640). Regulates osteoclastogenesis by adjusting the expression of CEBPB isoforms (By similarity). Plays an important regulatory role in the circadian clock function; regulates period length and rhythm amplitude of the suprachiasmatic nucleus (SCN) and liver clocks (By similarity).

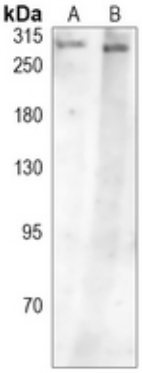
Cellular Location

Lysosome membrane; Peripheral membrane protein; Cytoplasmic side. Endoplasmic reticulum membrane; Peripheral membrane protein; Cytoplasmic side. Golgi apparatus membrane; Peripheral membrane protein; Cytoplasmic side. Cell membrane; Peripheral membrane protein. Mitochondrion outer membrane; Peripheral membrane protein; Cytoplasmic side. Cytoplasm. Nucleus {ECO:0000250|UniProtKB:Q9JLN9}. Nucleus, PML body {ECO:0000250|UniProtKB:Q9JLN9}. Microsome membrane. Cytoplasmic vesicle, phagosome. Note=Shuttles between cytoplasm and nucleus. Accumulates in the nucleus in response to hypoxia (By similarity). Targeting to lysosomes depends on amino acid availability and RRAGA and RRAGB (PubMed:18497260, PubMed:20381137). Lysosome targeting also depends on interaction with MEAK7. Translocates to the lysosome membrane in the presence of TM4SF5 (PubMed:30956113). The mTORC2 complex localizes to membranes: mTORC2 is active at the plasma membrane, endoplasmic reticulum membrane and lysosomes (PubMed:21867682). {ECO:0000250|UniProtKB:Q9JLN9, ECO:0000269|PubMed:18497260,

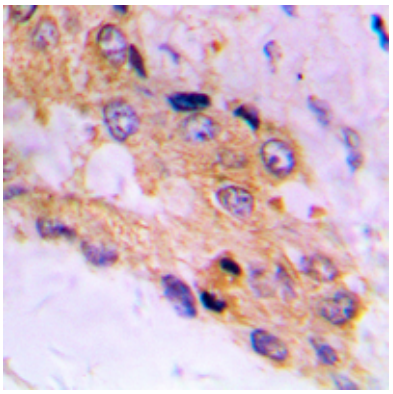
ECO:0000269|PubMed:20381137, ECO:0000269|PubMed:21867682, ECO:0000269|PubMed:29750193, ECO:0000269|PubMed:30956113}

Tissue Location Expressed in numerous tissues, with highest levels in testis.

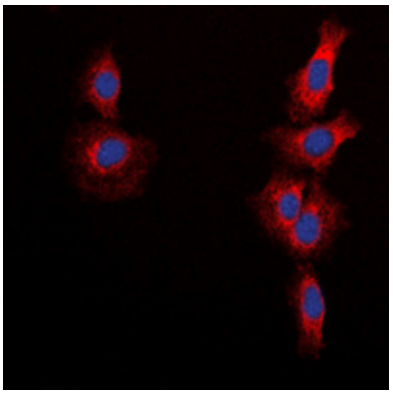
Images



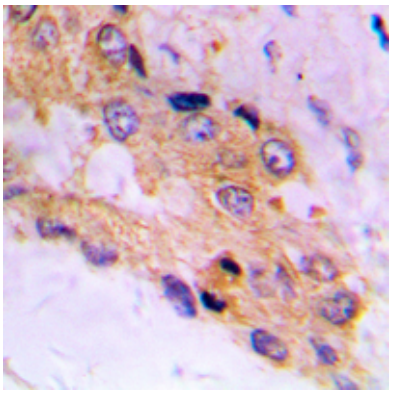
Western blot analysis of mTOR (Phospho-S2448) expression in U2OS (A), LO2 (B) whole cell lysates. (Predicted band size: 288 kD; Observed band size: 289 kD)



Immunohistochemical analysis of mTOR (Phospho-S2448) staining in human prostate cancer formalin fixed paraffin embedded tissue section. The section was pre-treated using heat mediated antigen retrieval with sodium citrate buffer (pH 6.0). The section was then incubated with the antibody at room temperature and detected using an HRP conjugated compact polymer system. DAB was used as the chromogen. The section was then counterstained with haematoxylin and mounted with DPX.



Immunofluorescent analysis of mTOR (Phospho-S2448) staining in MCF7 cells. Formalin-fixed cells were permeabilized with 0.1% Triton X-100 in TBS for 5-10 minutes and blocked with 3% BSA-PBS for 30 minutes at room temperature. Cells were probed with the primary antibody in 3% BSA-PBS and incubated overnight at 4 °C in a hidified chamber. Cells were washed with PBST and incubated with a DyLight 594-conjugated secondary antibody (red) in PBS at room temperature in the dark. DAPI was used to stain the cell nuclei (blue).



Immunohistochemical analysis of mTOR (pS2448) staining in human prostate cancer formalin fixed paraffin embedded tissue section. The section was pre-treated using heat mediated antigen retrieval with sodium citrate buffer (pH 6.0). The section was then incubated with the antibody at room temperature and detected using an HRP conjugated compact polymer system. DAB was used as the chromogen. The section was then counterstained with haematoxylin and mounted with DPX.

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