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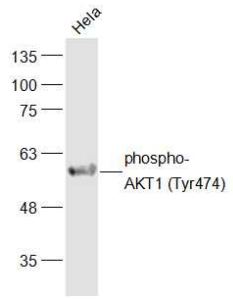
phospho-AKT1 (Tyr474) Rabbit pAb

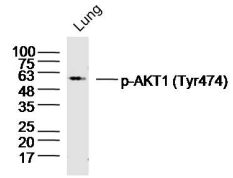
Catalog Number:	bs-10133R
Target Protein:	phospho-AKT1 (Tyr474)
Concentration:	1mg/ml
Form:	Liquid
Host:	Rabbit
Clonality:	Polyclonal
Isotype:	IgG
Applications:	WB (1:500-2000), IHC-P (1:100-500), IHC-F (1:100-500), IF (1:100-500), Flow-Cyt (1μg/Test), ICC/IF (1:100)
Reactivity:	Human, Mouse (predicted:Rat, Rabbit, Pig, Sheep, Cow, Chicken, Dog)
Predicted MW:	56 kDa
Entrez Gene:	207
Swiss Prot:	P31749
Source:	KLH conjugated synthesised phosphopeptide derived from human AKT1 around the phosphorylation site of Tyr474: FS(p-Y)SA.
Purification:	affinity purified by Protein A
Storage:	0.01M TBS (pH7.4) with 1% BSA, 0.02% Proclin300 and 50% Glycerol. Shipped at 4°C. Store at -20°C for one year. Avoid repeated freeze/thaw cycles.
Background:	This gene encodes one of the three members of the human AKT serine-threonine protein kinase family which are often referred to as protein kinase B alpha, beta, and gamma. These highly similar AKT proteins all have an N-terminal pleckstrin homology domain, a serine/threonine-specific kinase domain and a C-terminal regulatory domain. These proteins are phosphorylated by phosphoinositide 3-kinase (PI3K). AKT/PI3K forms a key component of many signalling pathways that involve the binding of membrane-bound ligands such as receptor tyrosine kinases, G-protein coupled receptors, and integrin-linked kinase. These AKT proteins therefore regulate a wide variety of cellular functions including cell proliferation, survival, metabolism, and angiogenesis in both normal and malignant cells. AKT proteins are recruited to the cell membrane by phosphatidylinositol 3,4,5-trisphosphate (PIP3) after phosphorylation of phosphatidylinositol 4,5-bisphosphate (PIP2) by PI3K. Subsequent phosphorylation of both threonine residue 308 and serine residue 473 is required for full activation of the AKT1 protein encoded by this gene. Phosphorylation of additional residues also occurs, for example, in response to insulin growth factor-1 and epidermal growth factor. Protein phosphatases act as negative regulators of AKT proteins by

dephosphorylating AKT or PIP3. The PI3K/AKT signalling pathway is crucial for tumor cell survival. Survival factors can suppress apoptosis in a transcription-independent manner by activating AKT1 which then phosphorylates and inactivates components of the apoptotic machinery. AKT proteins also participate in the mammalian target of rapamycin (mTOR) signalling pathway which controls the assembly of the eukaryotic translation initiation factor 4F (eIF4E) complex and this pathway, in addition to responding to extracellular signals from growth factors and cytokines, is disregulated in many cancers. Mutations in this gene are associated with multiple types of cancer and excessive tissue growth including Proteus syndrome and Cowden syndrome 6, and breast, colorectal, and ovarian cancers. Multiple alternatively spliced transcript variants have been found for this gene. [provided by RefSeq, Jul 2020]

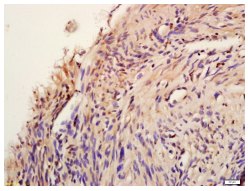
VALIDATION IMAGES



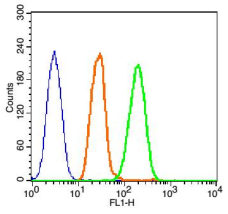
Sample: Hela(Human) Cell Lysate at 30 ug Primary: Anti-phospho-AKT1 (Tyr474) (bs-10133R) at 1/300 dilution Secondary: IRDye800CW Goat Anti-Rabbit IgG at 1/20000 dilution Predicted band size: 56 kD Observed band size: 56 kD



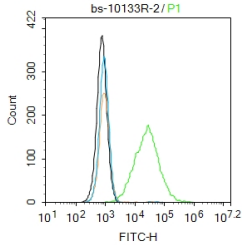
Sample: Lung (Mouse) Lysate at 40 ug Primary: Anti-phospho-AKT1 (Tyr474) (bs-10133R) at 1/300 dilution Secondary: IRDye800CW Goat Anti-Rabbit IgG at 1/20000 dilution Predicted band size: 56 kD Observed band size: 56 kD



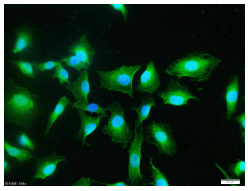
Tissue/cell: human myometrium tissue; 4% Paraformaldehyde-fixed and paraffin-embedded; Antigen retrieval: citrate buffer (0.01M, pH 6.0), Boiling bathing for 15min; Block endogenous peroxidase by 3% Hydrogen peroxide for 30min; Blocking buffer (normal goat serum,C-0005) at 37°C for 20 min; Incubation: Anti-phospho-AKT1(Tyr474) Polyclonal Antibody, Unconjugated(bs-10133R) 1:200, overnight at 4°C, followed by conjugation to the secondary antibody(SP-0023) and DAB(C-0010) staining



Primary Antibody:Rabbit Anti-phospho-AKT1 (Tyr474) antibody Supplier catalog number: bs-10133R (Green) Isotype Control Antibody: Rabbit IgG(orange) Secondary Antibody: Goat anti-rabbit IgG-FITC Positive control: Hela Cells(blue)



Blank control:A549. Primary Antibody (green line): Rabbit Anti-phospho-AKT1 (Tyr474) antibody (bs-10133R) Dilution: 2µg /10^6 cells; Isotype Control Antibody (orange line): Rabbit IgG . Secondary Antibody : Goat anti-rabbit IgG-FITC Dilution: 1µg /test. Protocol The cells were incubated in 5%BSA to block non-specific protein-protein interactions for 30 min 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.



Tissue/cell: A549 cell; 4% Paraformaldehyde-fixed; Triton X-100 at room temperature for 20 min; Blocking buffer (normal goat serum, C-0005) at 37°C for 20 min; Antibody incubation with (phospho-AKT1 (Tyr474)) polyclonal Antibody, Unconjugated (bs-10133R) 1:100, 90 minutes at 37°C; followed by a FITC conjugated Goat Anti-Rabbit IgG antibody at 37°C for 90 minutes, DAPI (blue, C02-04002) was used to stain the cell nuclei.

PRODUCT SPECIFIC PUBLICATIONS

[IF=5.561] Xinyang Fan. et al. CEBPA-Regulated Expression of SOCS1 Suppresses Milk Protein Synthesis through mTOR and JAK2-STAT5 Signaling Pathways in Buffalo Mammary Epithelial Cells. FOODS. 2023 Jan;12(4):708 WB ; Bovine . 36832783

[IF=5.223] Xinyang Fan. et al. MiR-190a regulates milk protein biosynthesis through the mTOR and JAK2–STAT5 signaling pathways by targeting PTHLH in buffalo mammary epithelial cells. J FUNCT FOODS. 2023 Mar;102:105451 WB ; Bovine . 10.1016/j.jff.2023.105451

[IF=2.8] Xianrong Xiong. et al. Maternal Kdm2a-mediated PI3K/Akt signaling and E-cadherin stimulate the morula-to-blastocyst transition revealing crucial roles in early embryonic development. THERIOGENOLOGY. 2023 Oct;209:60 WB ; Mouse . 37356280