– DATASHEET –

[Primary Antibody]

phospho-E2F1 (Ser332) Rabbit pAb



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- DATASHEET		1
Host: Rabbit Clonality: Polyclonal	Isotype: IgG	Applications: WB (1:500-2000) IHC-P (1:100-500)
GenelD: 1869	SWISS: Q01094	IHC-F (1:100-500) IF (1:100-500) Flow-Cyt (1ug/Test)
Target: phospho-E2F1 (Ser332)		Flow-Cyt (lug/lest)
Immunogen: KLH conjugated Synthesised phosphopeptide derived from human E2F1 around the phosphorylation site of Ser332: IV(p-S)PP.		Reactivity: Human, Mouse, Rat
Purification: affinity purif	ed by Protein A	
Concentration: 1mg/ml		Prodicted
Storage: 0.01M TBS (pH7.4) with 1% BSA, 0.02% Proclin300 and 50% Glycerol.		Predicted MW.: ^{46 kDa}
Shipped at 4°C. Store at -20°C for one year. Avoid repeated freeze/thaw cycles.		Subcellular Location: Nucleus
Background: E2F's are DNA binding proteins, which associate with negative regulators, such as the retinoblastoma p107 protein, resulting in an altered rate of gene transcription. The E2F proteins contain several evolutionally conserved domains found in most members of the family. These domains include a DNA binding domain, a dimerization domain which determines interaction with the differentiation regulated transcription factor proteins (DP), a transactivation domain enriched in acidic amino acids, and a tumor suppressor protein association domain which is embedded within the transactivation domain. This protein and another 2 members, E2F2 and E2F3, have an additional cyclin binding domain. E2F1 is proposed to be involved in several cellular processes that range from tumor suppressor, cell progression and oncogenesis. E2F1 overexpression can also drive cells into apoptosis.		
- VALIDATION IMAGES		



Sample: Lane 1: Human Hela cell lysates Lane 2: Human A673 cell lysates Lane 3: Human Molt-4 cell lysates Lane 4: Human U251cell lysates Lane 5: Human HepG2 cell lysates Primary: Antiphospho-E2F1 (Ser332) (bs-5306R) at 1/1000 dilution Secondary: IRDye800CW Goat Anti-Rabbit IgG at 1/20000 dilution Predicted band size: 46 kDa Observed band size: 61 kDa



Tissue/cell: human skin tissue; 4% Paraformaldehyde-fixed and paraffinembedded; 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-E2F1(Ser332) Polyclonal Antibody, Unconjugated(bs-5306R) 1:500, overnight at 4°C, followed by conjugation to the secondary antibody(SP-0023) and DAB(C-0010) staining



Tissue/cell: human colon cancer; 4% Paraformaldehyde-fixed and paraffinembedded; 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-E2F1(Ser332) Polyclonal Antibody, Unconjugated(bs-5306R) 1:500, overnight at 4°C, followed by conjugation to the secondary antibody(SP-0023) and DAB(C-0010) staining



Blank control (Black line):Molt4 (Black). Primary Antibody (green line): Rabbit Anti-phospho-E2F1 (Ser332) antibody (bs-5306R) Dilution: 1µg /10^6 cells; Isotype Control Antibody (orange line): Rabbit IgG . Secondary Antibody (white blue line): Goat anti-rabbit IgG-AF647 Dilution: 1µg /test. Protocol The cells were fixed with 4% PFA (10min at room temperature) and then permeabilized with 90% ice-cold methanol 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 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.