Phospho-AKT1 (Ser473) Monoclonal Antibody (SDRNR), APC, eBioscience™

Product Details	
Size	100 Tests
Species Reactivity	Human, Mouse
Published Species	Dog, Mouse, Human
Host/Isotype	Mouse / IgG2a, kappa
Recommended Isotype Control	Mouse IgG2a kappa Isotype Control (eBM2a), APC, eBioscience™
Class	Monoclonal
Type	Antibody
Clone	SDRNR
Conjugate	APC
Excitation/Emission Max	651/660 nm
Form	Liquid
Concentration	5 µL/Test
Purification	Affinity chromatography
Storage buffer	PBS, pH 7.2, with 0.2% BSA
Contains	0.09% sodium azide
Storage conditions	4° C, store in dark, DO NOT FREEZE!
RRID	AB_2573310

Applications	Tested Dilution	Publications
Flow Cytometry (Flow)	5 µL (0.5 µg)/test	14 Publications

Product Specific Information

Description: This SDRNR monoclonal antibody recognizes human and mouse AKT (also known as Protein Kinase B (PKB)) when phosphorylated on S473. AKT is a serine/threonine protein kinase that plays a key role in multiple cellular processes including metabolism, proliferation, apoptosis/survival, and migration. There are three homologous isoforms of AKT: AKT1, AKT2, and AKT3. AKT is activated by binding of its pleckstrin homology (PH) domain to membrane phospholipids and by phosphorylation. Phosphorylation of AKT at T308 by PDK1 and at S473 is required for full activation of this kinase. AKT promotes cell survival by inhibiting apoptosis via phosphorylation and inactivation of several targets including Bad, Foxo1, c-Raf, and caspase-9. Deregulation of AKT has been implicated as a major contributing factor in many types of cancer. AKT is negatively regulated by the phosphatase PTEN as well as by the chemical inhibitor LY294002. Specificity of this SDRNR clone was determined by ELISA, flow cytometry, and western blotting.

Applications Reported: This SDRNR antibody has been reported for use in intracellular staining followed by flow cytometric analysis.

Applications Tested: This SDRNR antibody has been pre-titrated and tested by intracellular staining followed by flow cytometric analysis of normal human peripheral blood cells. This can be used at 5 µL (0.5 µg) per test. A test is defined as the amount (µg) of antibody that will stain a cell sample in a final volume of 100 µL. Cell number should be determined empirically but can range from 10^5 to 10^8 cells/test.

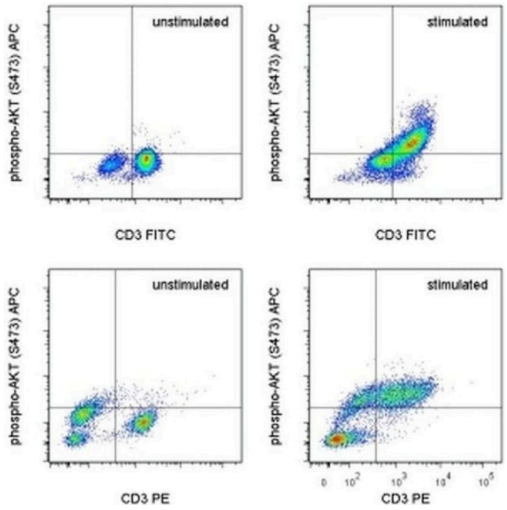
Staining Protocol: All protocols work well for this monoclonal antibody. Use of Protocol A: Two-step protocol: intracellular

(cytoplasmic) proteins allows for the greatest flexibility for detection of surface and intracellular (cytoplasmic) proteins. Use of Protocol B: One-step protocol: intracellular (nuclear) proteins is recommended for staining of transcription factors in conjunction with surface and phosphorylated intracellular (cytoplasmic) proteins. Protocol C: Two-step protocol: Fixation /Methanol allows for the greatest discrimination of phospho-specific signaling between unstimulated and stimulated samples, but with limitations on the ability to stain specific surface proteins (refer to "Clone Performance Following Fixation /Permeabilization" located in the Best Protocols Section under the Resources tab online). All Protocols can be found in the Flow Cytometry Protocols: "Staining Intracellular Antigens for Flow Cytometry Protocol" located in the Best Protocols Section under the Resources tab online.

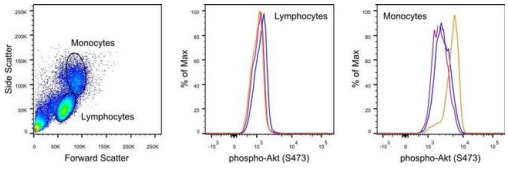
Excitation: 633-647 nm; Emission: 660 nm; Laser: Red Laser.

Filtration: 0.2 µm post-manufacturing filtered.

Product Images For Phospho-AKT1 (Ser473) Monoclonal Antibody (SDRNR), APC, eBioscience™



Phospho-AKT1 (Ser473) Antibody (17-9715-42) in Flow
TOP: Normal human peripheral blood cells were unstimulated (left) or stimulated with Anti-Human CD3 Functional Grade Purified (Product # 16-0037-81) and Anti-Human CD28 Functional Grade Purified (Product # 16-0289-81) for 48 hours (right). The cells were then intracellularly stained with Anti-Human CD3 FITC (Product # 11-0036-42) and Anti-Human/Mouse phospho-AKT (S473) APC using the Intracellular Fixation & Permeabilization Buffer Set (Product # 88-8824-00) and protocol. Cells in the lymphocyte gate were used for analysis. BOTTOM: Mouse splenocytes were unstimulated (left) or stimulated with Anti-Mouse CD3e Functional Grade Purified (Product # 16-0031-82) and Anti-Mouse CD28 Functional Grade Purified (Product # 16-0281-82) for 48 hours (right). The cells were then intracellularly stained with Anti-Mouse CD3e PE (Product # 12-0031-82) and Anti-Human/Mouse phospho-AKT (S473) APC using the Intracellular Fixation & Permeabilization Buffer Set (Product # 88-8824-00) and protocol. Cells in the lymphocyte gate were used for analysis.



Phospho-AKT1 (Ser473) Antibody (17-9715-42)
Intracellular staining of stimulated human peripheral blood cells. As expected based on known expression patterns, phospho-AKT (S473) clone SDRNR stains monocytes in response to LPS (right) treatment but does not stain lymphocytes (middle). Details: Normal human peripheral blood cells were unstimulated (purple histogram) or stimulated with Lipopolysaccharide (LPS) Solution (500X) (orange histogram), then intracellularly stained with Anti-Human/Mouse phospho-AKT (S473) (clone SDRNR) using the Intracellular Fixation and Permeabilization Buffer Set and protocol. Single cells in the lymphocyte (middle) and monocyte (right) gates were used for analysis. Staining of the isotype control, Mouse IgG2a kappa, on LPS-stimulated cells is shown in both plots (blue histogram). {TM}

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14 References

Flow Cytometry (14)

Biomarker insights

Fc Receptor-Like 1 as a Promising Target for Immunotherapeutic Interventions of B-Cell-Related Disorders.

"Published figure using Phospho-AKT1 (Ser473) monoclonal antibody (Product # 17-9715-42) in Flow Cytometry"

Authors: Yousefi Z,Sharifzadeh S,Yar-Ahmadi V,Andalib A,Eskandari N

Year
2023

Nature

Neonatal imprinting of alveolar macrophages via neutrophil-derived 12-HETE.

"17-9715-42 was used in Flow cytometry/Cell sorting to highlight the complexity of prenatal RTM programming and reveal their dependency on in trans eicosanoid production by neutrophils for lifelong self-renewal."

Authors: Pernet E,Sun S,Sarden N,Gona S,Nguyen A,Khan N,Mawhinney M,Tran KA,Chronopoulos J,Amberkar D, Sadeghi M,Grant A,Wali S,Prevel R,Ding J,Martin JG,Thanabalasuriar A,Yipp BG,Barreiro LB,Divangahi M

Year
2023

Species
Mouse
Dog

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