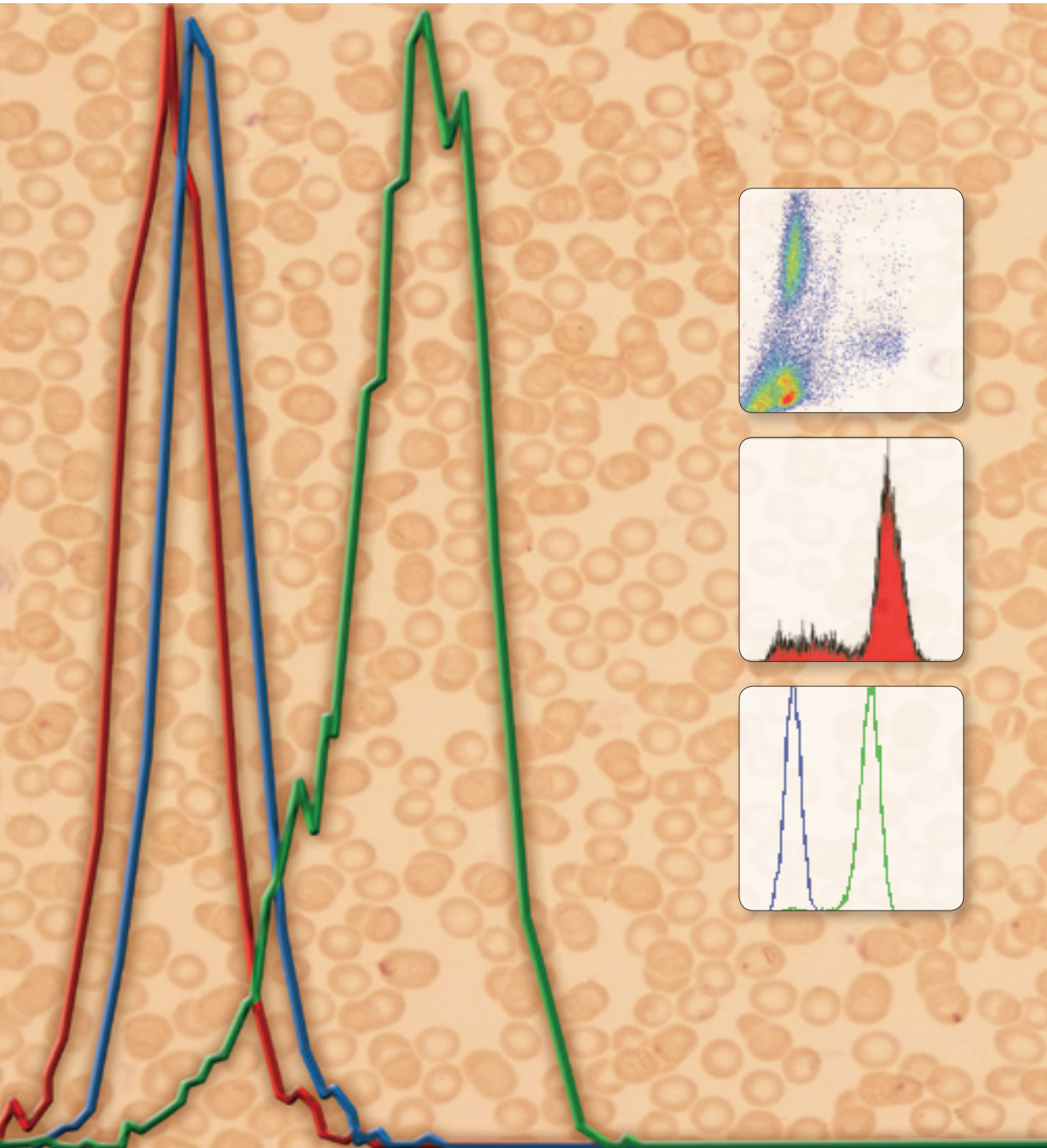


ANTIBODIES, KITS, AND REAGENTS FOR FLOW CYTOMETRY

Unparalleled Product Quality, Validation, and Technical Support



Cell Signaling

TECHNOLOGY®

XP[®] Monoclonal Antibodies

one antibody, multiple applications[™]

XP[®] monoclonal antibodies are a line of high quality rabbit monoclonal antibodies exclusively available from Cell Signaling Technology (CST). Any product labeled with XP has been carefully selected based on superior performance in the most relevant research applications.

These antibodies are generated using XMT[®] technology, a proprietary monoclonal method developed at CST. The technology provides access to a broad range of antibodies unattainable with traditional monoclonal technologies, allowing more comprehensive screening and the identification of XP monoclonal antibodies with exceptional specificity, sensitivity, and performance.

eXceptional specificity

As with all of our antibodies, the antibody is specific to your target of interest, saving you valuable time and resources.

+ eXceptional sensitivity

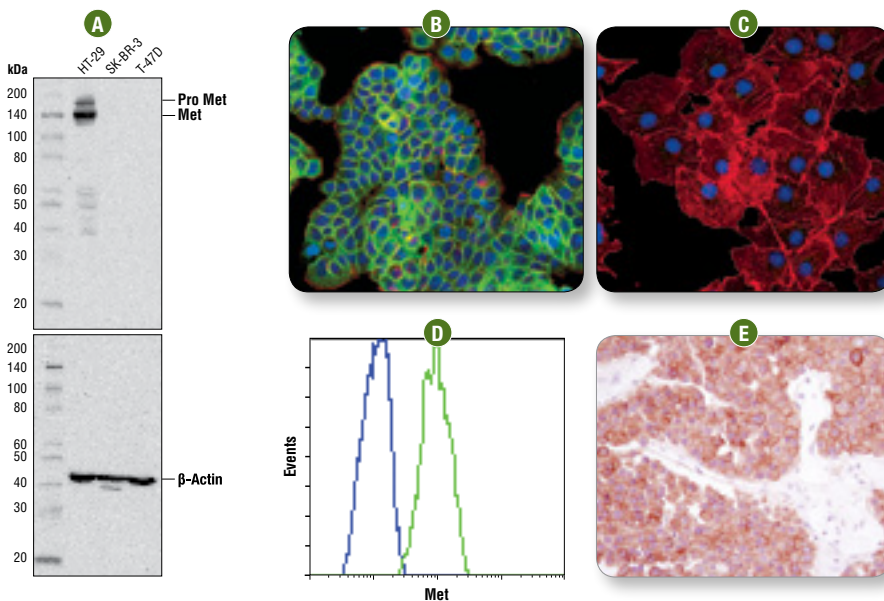
The antibody will provide a stronger signal for your target protein in cells and tissues, allowing you to monitor expression of low levels of endogenous proteins, saving you valuable materials.

+ eXceptional stability and reproducibility

XMT technology combined with our stringent quality control ensures maximum lot-to-lot consistency and the most reproducible results.

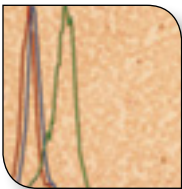
= eXceptional Performance[™]

XMT technology coupled with our extensive antibody validation and stringent quality control delivers XP monoclonal antibodies with eXceptional Performance in the widest range of applications.



Met (D1C2) XP[®] Rabbit mAb #8198: Western blot analysis (A) of extracts from HT-29 (Met+), SK-BR-3 (Met-), and T-47D (Met-) cells using #8198 (upper) or β -Actin Antibody #4967 (lower). Confocal IF analysis of HT-29 (B) and T-47D (C) cells using #8198 (green). Actin filaments were labeled with DY-554 phalloidin (red). Blue pseudocolor = DRAQ5[®] #4084 (fluorescent DNA dye). Flow cytometric analysis (D) of T-47D (blue) and HT-29 (green) cells using #8198. IHC analysis (E) of paraffin-embedded human hepatocellular carcinoma using #8198.

Visit our website for more experimental details, additional information, and a complete list of available XP[®] monoclonal antibodies.



Please see page 4
for full details about
antibody validation for
flow cytometry.



WINNER!

Life Science
Industry Awards®

■ **Best Antibodies**

■ **Best Breakthrough Products
for Cancer Research**

Application Key:

W Western / **IP** Immunoprecipitation / **IHC** Immunohistochemistry
IF Immunofluorescence / **F** Flow Cytometry / **ChIP** Chromatin
Immunoprecipitation / **(-IC)** Immunocytochemistry, **-P** Paraffin,
-F Frozen / **E-P** Peptide ELISA

Reactivity Key:

H human / **M** mouse / **R** rat / **Hm** hamster / **Mk** monkey
C chicken / **Mi** mink / **Dm** D. melanogaster / **X** Xenopus
Z zebra fish / **B** bovine / **Dg** dog / **Pg** pig / **Sc** S. cerevisiae
All all species expected / () 100% sequence homology

For Research Use Only. Not For Use In Diagnostic Procedures.

Antibodies and Reagents for Flow Cytometry

Cell Signaling Technology (CST) provides the highest quality activation state and total protein antibodies available for use in Flow Cytometry research. Our in-house Flow Cytometry Group has validated each Flow Cytometry recommended CST™ antibody using multiple approaches. Technical support is provided by the same scientists who validate the products.

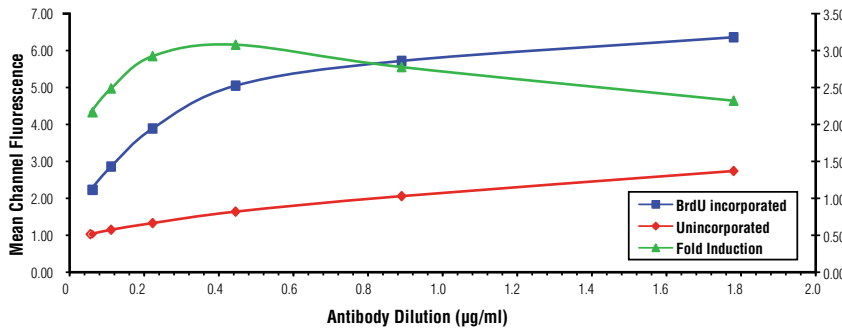
- 4 | Antibody Validation for Flow Cytometry
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- 14 | Cell Cycle, Checkpoint Control, and DNA Damage
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- 25 | Other Antibodies
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Antibody Validation for Flow Cytometry

Cell Signaling Technology (CST) antibodies have undergone rigorous validation by our in-house Flow Cytometry Group. Our goal is to provide our customers with the most specific antibodies that yield the brightest signal with the lowest background possible. All of our 550+ antibodies validated for flow cytometry research have been screened to determine optimal dilution and to verify specificity.

Titration:

Serial dilution is used to determine optimal dilution.

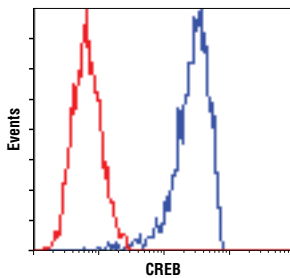


BrdU (Bu20a) Mouse mAb #5292:

Flow cytometric analysis of Jurkat cells, unincorporated (red) or after 30 min of BrdU incorporation (blue), using serial dilutions of #5292. The fold-induction ratio is shown in green. Optimal concentration of #5292 was determined to be 0.044 µg/ml.

Isotype Control:

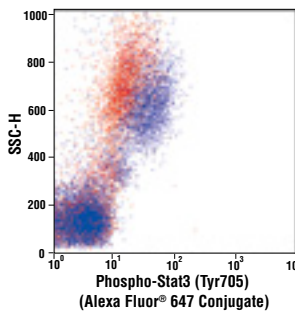
Comparison of signal to isotype control is used to estimate the nonspecific binding of primary antibodies.



Rabbit (DA1E) mAb IgG XP® Isotype Control #3900: Flow cytometric analysis of SH-SY5Y cells using CREB (D76D11) Rabbit mAb Antibody #4820 (blue) compared to concentration matched #3900 (red).

Activation-state Specificity:

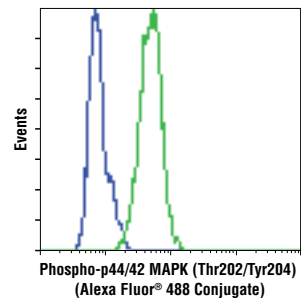
Comparison of the signal on treated and untreated cells helps to verify activation-state specificity.



Phospho-Stat3 (Tyr705) (D3A7) XP® Rabbit mAb (Alexa Fluor® 647 Conjugate) #4324: Flow cytometric analysis of human whole blood, untreated (red) or treated with Human Granulocyte Colony Stimulating Factor (hG-CSF) #8930 (blue), using #4324.

Inhibitor Treatment:

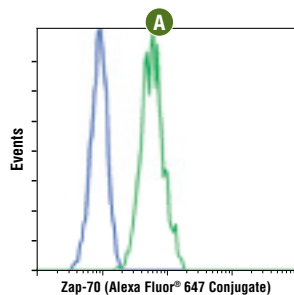
Treatment of cell lines with pathway-specific inhibitors helps to verify target specificity.



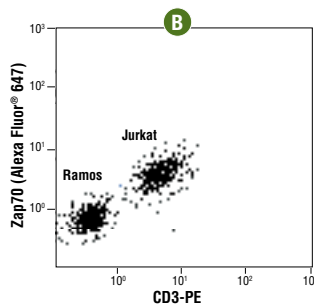
Phospho-p44/42 MAPK (Erk1/2) (Thr202/Tyr204) (D13.14.4E) XP® Rabbit mAb (Alexa Fluor® 488 Conjugate) #4344: Flow cytometric analysis of Jurkat cells treated with U0126 #9903 (blue) or TPA #4174 (green), using #4344.

Positive and Negative Cell Lines:

Use of known positive and negative cell lines verifies target specificity.



Zap-70 (136F12) Rabbit mAb (Alexa Fluor® 647 Conjugate) #2707: Flow cytometric analysis (A) of Ramos B (blue) and Jurkat T (green) cells using #2707. Two-color flow cytometric analysis (B) of a mixed population of T and B cells (Jurkat and Ramos, respectively) using #2707 and a CD3 antibody. CD3-negative B cells have little or no Zap-70 staining, while CD3-positive T cells stain brightly for Zap-70 protein.



Other Validation Steps Include:

- ❑ The use of blocking peptides, siRNA, and expression vectors verifies specificity of staining.
- ❑ Phosphatase treatment confirms phospho-specificity of the antibody.
- ❑ Extensive quality control testing guarantees stability over time and eliminates lot-to-lot variability.
- ❑ Optimized protocols are provided and dilutions are predetermined.
- ❑ Concurrent testing on other relevant applications.

Complementary Reagents

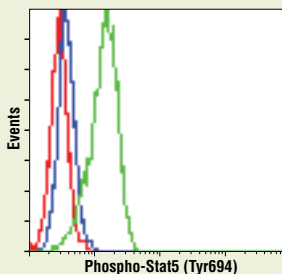
CST offers a wide selection of conjugated secondary antibodies and cellular dyes, as well as isotype controls. These same reagents are also used in-house for antibody validation in flow cytometry analysis and therefore work optimally with our primary antibodies. Our research protocols (included in the back of this brochure) also recommend the use of these reagents. Technical support is provided by the product scientists who make the reagents and know them best.

Isotype Controls

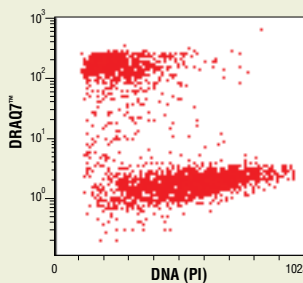
	#5415 Mouse (G3A1) mAb IgG1 Isotype Control
New	#6899 Mouse (G3A1) mAb IgG1 Isotype Control (PE Conjugate)
	#4878 Mouse (MOPC-21) mAb IgG1 Isotype Control (Alexa Fluor® 488 Conjugate)
	#4843 Mouse (MOPC-21) mAb IgG1 Isotype Control (Alexa Fluor® 647 Conjugate)
	#4097 Mouse (MOPC-21) mAb IgG1 Isotype Control (Biotinylated)
XP	#3900 Rabbit (DA1E) mAb IgG XP® Isotype Control
XP	#2975 Rabbit (DA1E) mAb IgG XP® Isotype Control (Alexa Fluor® 488 Conjugate)
XP	#2985 Rabbit (DA1E) mAb IgG XP® Isotype Control (Alexa Fluor® 647 Conjugate)
XP	#4096 Rabbit (DA1E) mAb IgG XP® Isotype Control (Biotinylated)
XP	#5742 Rabbit (DA1E) mAb IgG XP® Isotype Control (PE Conjugate)
	#4340 Rabbit IgG Isotype Control (Alexa Fluor® 488 Conjugate)
	#3452 Rabbit IgG Isotype Control (Alexa Fluor® 647 Conjugate)

Product References:

Rabbit IgG Isotype Control (Alexa Fluor® 488 Conjugate) #4340: Anand, S. et al. (2011) Increased basal intracellular signaling patterns do not correlate with JAK2 genotype in human myeloproliferative neoplasms. *Blood*. 118, 1610-1621.



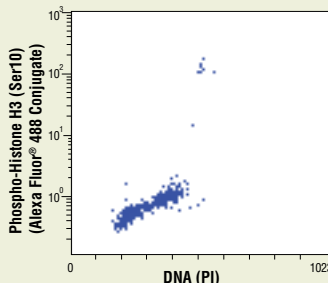
Rabbit (DA1E) mAb IgG XP® Isotype Control #3900: Flow cytometric analysis of K-562 cells, untreated (green) or treated with imatinib #9084 (blue), using Phospho-Stat5 (Tyr694) (C71E5) Rabbit mAb #9314 compared to concentration-matched #3900 (red).



DRAQ7™ #7406: Flow cytometric analysis of live (unpermeabilized) Jurkat cells treated with Staurosporine #9953. Gated population represents DRAQ7™-positive apoptotic cells.

Dyes and Secondary Antibodies

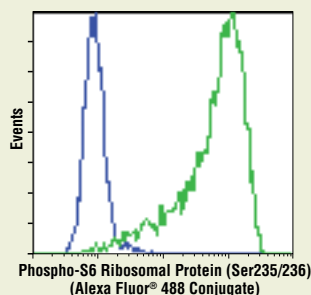
	#4408 Anti-mouse IgG (H+L), F(ab') ₂ Fragment (Alexa Fluor® 488 Conjugate)
	#4410 Anti-mouse IgG (H+L), F(ab') ₂ Fragment (Alexa Fluor® 647 Conjugate)
	#4412 Anti-rabbit IgG (H+L), F(ab') ₂ Fragment (Alexa Fluor® 488 Conjugate)
	#4414 Anti-rabbit IgG (H+L), F(ab') ₂ Fragment (Alexa Fluor® 647 Conjugate)
	#4416 Anti-rat IgG (H+L), (Alexa Fluor® 488 Conjugate)
	#4418 Anti-rat IgG (H+L), (Alexa Fluor® 647 Conjugate)
	#4084 DRAQ5®
New	#7406 DRAQ7™
New	#4087 Propidium Iodide (PI)/ RNase Staining Solution



Propidium Iodide (PI)/RNase Staining Solution #4087: Flow cytometric analysis of Jurkat cells using Phospho-Histone H3 (Ser10) Antibody (Alexa Fluor® 488 Conjugate) #9708 and #4087.

Chemical Activators and Inhibitors

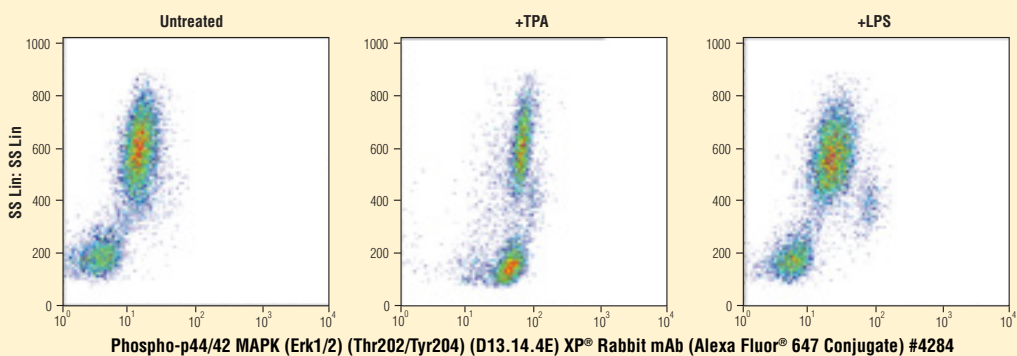
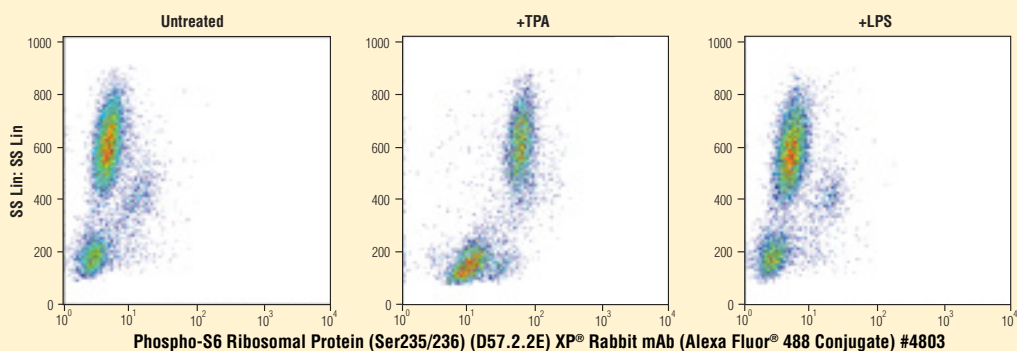
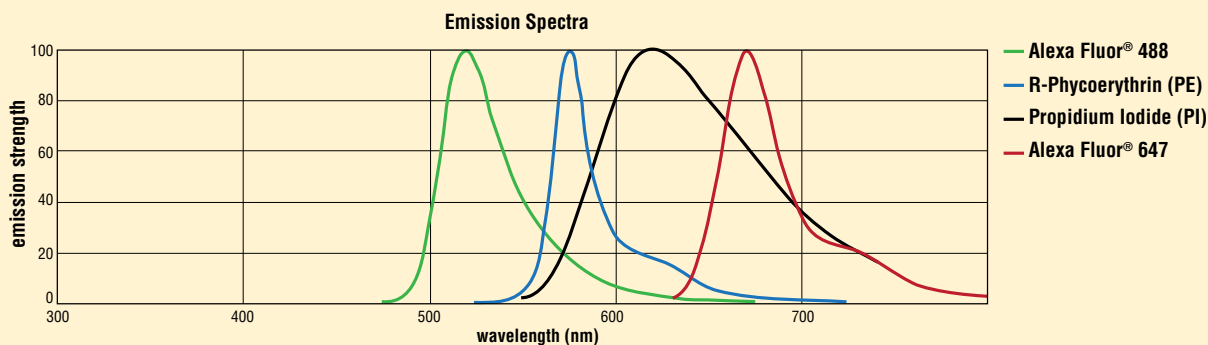
	#9944 AICAR	New	#8158 SB202190
	#9841 Bisindolylmaleimide I, Hydrochloride		#5633 SB203580 (p38 Inhibitor)
	#9972 Brefeldin A	New	#8177 SP600125
	#9902 Calyculin A (Serine/Threonine Phosphatase Inhibitor)		#9903 U0126 (MEK1/2 Inhibitor)
	#9973 Cyclosporin A		#9900 PD98059 (MEK1 Inhibitor)
New	#9052 Dasatinib		#9996 Oligomycin
	#9886 Docetaxel		#9807 Paclitaxel
	#9974 FK-506	New	#9493 PKC412
	#3828 Forskolin		#9904 Rapamycin (FRAP/mTOR Inhibitor)
	#9843 Geldanamycin		#9885 Roscovitine
	#9844 H-89, Dihydrochloride		#9953 Staurosporine
New	#9084 Imatinib		#4174 TPA (12-O-Tetradecanoylphorbol-13-Acetate)
	#9995 Ionomycin, Calcium Salt		#9950 Trichostatin A (TSA)
New	#9676 Leptomycin B		#9842 Tyrphostin AG 1478
	#9901 LY294002 (PI3 Kinase Inhibitor)		#9951 Wortmannin



LY294002 #9901, Wortmannin #9951, and U0126 #9903: Flow cytometric analysis of Jurkat cells, untreated (green) or treated with #9901, #9951, and #9903 (blue), using Phospho-S6 Ribosomal Protein (Ser235/236) (D57.2.2E) XP® Rabbit mAb (Alexa Fluor® 488 Conjugate) #4803.

Conjugated Antibodies

Cell Signaling Technology (CST) offers Alexa Fluor® and PE-conjugated antibodies that are optimized for flow cytometry research. The combination of Alexa Fluor® and PE dyes, which exhibit superior brightness and photostability, with the highest quality CST™ antibodies results in conjugates with bright signal and low background. All conjugated antibodies are validated in-house. Price-competitive and high-quality custom antibody conjugation of CST antibodies is offered from our Custom Conjugation Group, please see page 24 for additional information. Contact your sales representative with any inquiries.



Flow cytometric analysis of whole blood, either untreated, treated with TPA #4174, or LPS-treated, using either Phospho-S6 Ribosomal Protein (Ser235/236) (D57.2.2E) XP® Rabbit mAb (Alexa Fluor® 488 Conjugate) #4803 (top) or Phospho-p44/42 MAPK (Erk1/2) (Thr202/Tyr204) (D13.14.4E) XP® Rabbit mAb (Alexa Fluor® 647 Conjugate) #4284 (bottom).

Isotype Controls

	Reactivity	Conjugates		
		488	PE	647
Mouse IgG1 Isotype Control (MOPC-21)	–	#4878	–	#4843
XP #3900 Rabbit (DA1E) mAb IgG XP® Isotype Control	–	#2975	#5742	#2985
Rabbit IgG (Polyclonal) Isotype Control	–	#4340	–	#3452
#5415 Mouse (G3A1) mAb IgG1 Isotype Control	–	–	#6899	–

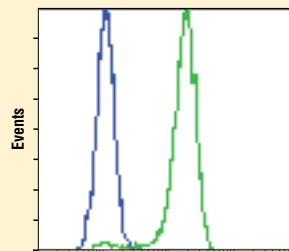
Epitope Tag Antibody

	Reactivity	488	PE	647
#2368 DYKDDDDK Tag Antibody (Binds to same epitope as Sigma's Anti-FLAG® M2 Antibody)	All	#5407	–	#3916
#2624 GST (26H1) Mouse mAb	All	#3368	–	#3445
#2367 HA-Tag (6E2) Mouse mAb	All	#2350	–	#3444
#2276 Myc-Tag (9B11) Mouse mAb	All	#2279	#3739	#2233

Unconjugated Antibody

	Reactivity	488	PE	647
#2855 Phospho-4E-BP1 (Thr37/46) (236B4) Rabbit mAb	H, M, R, Mk, Dm	#2846	#7547	#5123
#4970 β-Actin (13E5) Rabbit mAb	H, M, R, Mk, B, Pg, (C, Dg)	#8844	–	#8584
#2965 Phospho-Akt (Thr308) (C31E5E) Rabbit mAb	H, M, R, Hm, Mk	#2918	#9088	#3375
XP #4060 Phospho-Akt (Ser473) (D9E) XP® Rabbit mAb	H, M, R, Hm, Mk, Dm, Z, B, (C, X, Dg, Pg)	#4071	#5315	#4075
#4058 Phospho-Akt (Ser473) (193H12) Rabbit mAb	H, M, R	#2336	–	#2337
#4691 Akt (pan) (C67E7) Rabbit mAb	H, M, R, Mk, Dm	#5084	–	#5186
#2966 Akt (5G3) Mouse mAb	H, M, R, Hm	#2917	–	#2944
XP #5153 Androgen Receptor (D6F11) XP® Rabbit mAb	H	#7395	#8428	#7397
XP #2914 Phospho-Aurora A (Thr288)/Aurora B (Thr232)/Aurora C (Thr198) (D13A11) XP® Rabbit mAb	H, M, R	#8525	–	–
#2827 Phospho-Bcl-2 (Ser70) (5H2) Rabbit mAb	H	#2834	–	–
#2764 Bcl-xL (54H6) Rabbit mAb	H, M, R, Mk	#2767	–	–
#3195 E-Cadherin (24E10) Rabbit mAb	H, M, (Dg, Pg)	#3199	#7559	#9835
#9664 Cleaved Caspase-3 (Asp175) (5A1E) Rabbit mAb	H, M, R, Mk, (Dg)	–	#9978	–
#9661 Cleaved Caspase-3 (Asp175) Antibody	H, M, R, Mk, (B, Dg, Pg)	#9669	–	–
#2677 β-Catenin (L54E2) Mouse mAb (IF Preferred)	H, (M, R, Mk, Pg)	#2849	#6898	#4627
#3570 CD44 (156-3C11) Mouse mAb	H	#3516	#8724	–
#4850 COX IV (3E11) Rabbit mAb	H, R, Mk, Z, B, Pg	#4853	–	#7561
#9198 Phospho-CREB (Ser133) (87G3) Rabbit mAb	H, M, R	#9187	–	–
#4135 Cyclin B1 (V152) Mouse mAb	H, M, (Hm)	#4112	–	#4118
XP #4267 EGF Receptor (D38B1) XP® Rabbit mAb	H, M, Mk	#5616	#8839	#5588
#2929 EpCAM (VU1D9) Mouse mAb	H	#5198	#8995	#5447
#2251 FosB (5G4) Rabbit mAb	H, M, R	#2023	–	–
#2118 GAPDH (14C10) Rabbit mAb	H, M, R, Mk	#3906	–	#3907
#2024 Hexokinase I (C35C4) Rabbit mAb	H, M	#3689	–	#3540
#9718 Phospho-Histone H2A.X (Ser139) (20E3) Rabbit mAb	H, M, R, Mk	#9719	#5763	#9720
XP #3377 Phospho-Histone H3 (Ser10) (D2C8) XP® Rabbit mAb	H, M, R, Mk, Z	#3465	#5764	#3458
#9701 Phospho-Histone H3 (Ser10) Antibody	H, M, R, Mk, C, Dm, Z, Sc, (X)	#9708	–	#9716
#9649 Acetyl-Histone H3 (Lys9) (C5B11) Rabbit mAb	H, M, R, Mk, Z, (Sc)	#9683	–	#4484
#4876 HSP70 (D69) Antibody	H, M, R, Mk	#4837	–	#4008
#4814 IκBα (L35A5) Mouse mAb (Amino-terminal Antigen)	H, M, R, Mk, B, Pg	#5743	#7523	–
#4545 Pan-Keratin (C11) Mouse mAb	H, R, Mk	#4523	#5075	#4528
#3308 c-Kit (Ab81) Mouse mAb	H	#3310	–	–
#4777 Lamin A/C (4C11) Mouse mAb	H, M, R, Mk	#8617	–	–
#2230 LEF1 (C12A5) Rabbit mAb	H, M, R	#8490	–	–

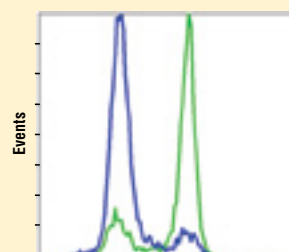
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Androgen Receptor (Alexa Fluor® 488 Conjugate)

Androgen Receptor (D6F11) XP® Rabbit mAb (Alexa Fluor® 488 Conjugate)

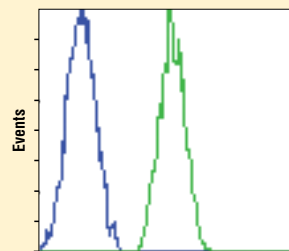
#7395: Flow cytometric analysis of Jurkat (blue) and LNCaP (green) cells using #7395.



Cleaved Caspase-3 (Alexa Fluor® 488)

Cleaved Caspase-3 (Asp175) Antibody (Alexa Fluor® 488 Conjugate) #9669:

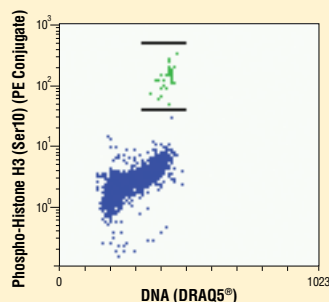
Flow cytometric analysis of Jurkat cells, untreated (blue) or etoposide-treated (green), using #9669.



EGF Receptor (Alexa Fluor® 647 Conjugate)

EGF Receptor (D38B1) XP® Rabbit mAb (Alexa Fluor® 647 Conjugate) #5588:

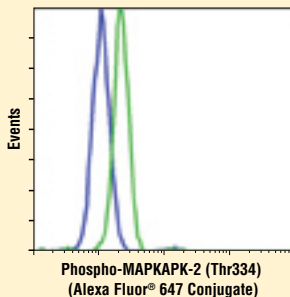
Flow cytometric analysis of Jurkat (blue) and HeLa (green) cells using #5588.



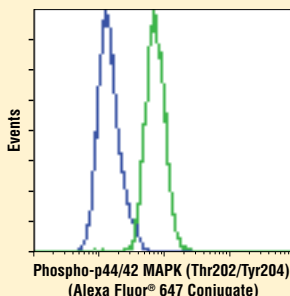
Phospho-Histone H3 (Ser10) (D2C8) XP® Rabbit mAb (PE Conjugate)

#5764: Flow cytometric analysis of Jurkat cells using #5764 and DRAQ5® #4084 (DNA content). Gated population represents phospho-histone H3-positive cells.

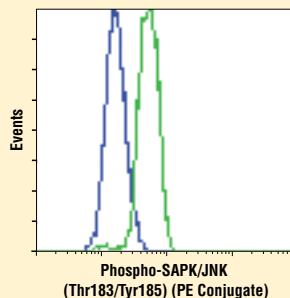
Conjugated Antibodies Continued



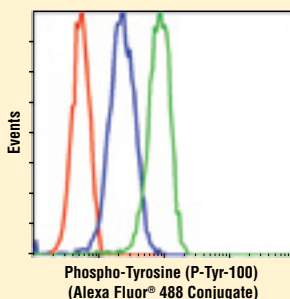
Phospho-MAPKAPK-2 (Thr334) (27B7) Rabbit mAb (Alexa Fluor® 647 Conjugate) #4320: Flow cytometric analysis of THP-1 cells, untreated (blue) or anisomycin-treated (green), using #4320.



Phospho-p44/42 MAPK (Erk1/2) (Thr202/Tyr204) (D13.14.4E) XP® Rabbit mAb (Alexa Fluor® 647 Conjugate) #4284: Flow cytometric analysis of Jurkat cells, treated with U0126 #9951 (blue) or TPA #4174 (green), using #4284.



Phospho-SAPK/JNK (Thr183/Tyr185) (G9) Mouse mAb (PE Conjugate) #5755: Flow cytometric analysis of THP-1 cells, untreated (blue) or anisomycin-treated (green), using #5755.



Phospho-Tyrosine Mouse mAb (P-Tyr-100) (Alexa Fluor® 488 Conjugate) #9414: Flow cytometric analysis of K-562 cells, untreated (green) or treated with imatinib #9084 (blue), using #9414 compared to a concentration-matched nonspecific negative control antibody (red).

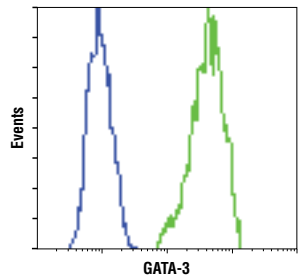
Unconjugated Antibody

	Reactivity	Conjugates		
		488	PE	647
#3007 Phospho-MAPKAPK-2 (Thr334) (27B7) Rabbit mAb	H, M, R, Mk	#4338	–	#4320
XP® #8198 Met (D1C2) XP® Rabbit mAb	H	#8494	–	–
#2983 mTOR (7C10) Rabbit mAb	H, M, R, Mk	#5043	–	#5048
XP® #4903 Nanog (D73G4) XP® Rabbit mAb	H, (Mk)	–	–	#5448
XP® #5482 Phospho-NDRG1 (Thr346) (D98G11) XP® Rabbit mAb	H, M, R, Mk	#6992	–	#7497
#3033 Phospho-NF-κB p65 (Ser536) (93H1) Rabbit mAb	H, M, R, Hm, Mk, Pg, (Dg)	#4886	#5733	#4887
#2840 Oct-4A (C30A3) Rabbit mAb	H, M	#5177	–	#5263
#2947 p21 Waf1/Cip1 (12D1) Rabbit mAb	H, Mk	#5487	#8865	#8587
#9215 Phospho-p38 MAPK (Thr180/Tyr182) (3D7) Rabbit mAb	H, M, R, Mk, Dm, Pg, Sc, (Hm, Mi, Z, B)	–	#6908	–
#9216 Phospho-p38 MAPK (Thr180/Tyr182) (28B10) Mouse mAb	H, M, R, Mk, Sc, (Z)	#4551	–	#4552
XP® #4370 Phospho-p44/42 MAPK (Erk1/2) (Thr202/Tyr204) (D13.14.4E) XP® Rabbit mAb	H, M, R, Hm, Mk, Mi, Dm, Z, B, Dg, Pg, Sc	#4344	#5682	#4284
#9106 Phospho-p44/42 MAPK (Erk1/2) (Thr202/Tyr204) (E10) Mouse mAb	H, M, R, Hm, Mk, Mi, Dm, Z, B, Pg	#4374	–	#4375
#4695 p44/42 MAPK (Erk1/2) (137F5) Rabbit mAb	H, M, R, Hm, Mk, Mi, Dm, Z, B, Dg, Pg, (C)	#4780	–	#5376
#9286 Phospho-p53 (Ser15) (16G8) Mouse mAb	H	#9235	–	#8695
#2527 p53 (7F5) Rabbit mAb	H, Mk	#5429	–	–
#2524 p53 (1C12) Mouse mAb	H, M, R, Mk	#2015	–	#2533
XP® #5625 Cleaved PARP (Asp214) (D64E10) XP® Rabbit mAb	H, Mk	#9148	#8978	#6087
#2258 PU.1 (9G7) Rabbit mAb	H, M, (Mk, Pg)	#2216	–	#2240
XP® #4858 Phospho-S6 Ribosomal Protein (Ser235/236) (D57.2.2E) XP® Rabbit mAb	H, M, R, Mk, Sc, (C)	#4803	#5316	#4851
#4856 Phospho-S6 Ribosomal Protein (Ser235/236) (2F9) Rabbit mAb	H, M, R, Mk	#4854	–	–
XP® #5364 Phospho-S6 Ribosomal Protein (Ser240/244) (D68F8) XP® Rabbit mAb	H, M, R, Mk	#5018	–	#5044
#2317 S6 Ribosomal Protein (54D2) Mouse mAb	H, M, R, Mk, Dm	#5317	–	#5548
#9255 Phospho-SAPK/JNK (Thr183/Tyr185) (G9) Mouse mAb	H, M, R, Hm, Sc	–	#5755	#9257
XP® #3579 Sox2 (D6D9) XP® Rabbit mAb	H, (Mk, B, Dg)	#5049	–	#5067
#4755 SSEA4 (MC813) Mouse mAb	H	–	–	#5836
#9167 Phospho-Stat1 (Tyr701) (58D6) Rabbit mAb	H, M	#9174	#8062	#8009
XP® #9145 Phospho-Stat3 (Tyr705) (D3A7) XP® Rabbit mAb	H, M, R, Mk, (Hm, B, Pg)	#4323	#8119	#4324
#9314 Phospho-Stat5 (Tyr694) (C71E5) Rabbit mAb	H, M, (R, Mk, B)	#3939	–	#9365
#2808 Survivin (71G4B7) Rabbit mAb	H, M, R	#2810	–	#2866
#2125 α-Tubulin (11H10) Rabbit mAb	H, M, R, Mk, Dm, B, (Dg)	#5063	–	#5046
#2128 β-Tubulin (9F3) Rabbit mAb	H, M, R, Mk, Z, B, (C)	#3623	#5867	#3624
#9411 Phospho-Tyrosine Mouse mAb (P-Tyr-100)	All	#9414	–	#9415
XP® #5741 Vimentin (D21H3) XP® Rabbit mAb	H, M, R, Mk	#9854	–	#9856
XP® #3165 Zap-70 (D1C10E) XP® Rabbit mAb	H, M	#9473	–	–
Zap-70 (136F12) Rabbit mAb	H	–	–	#2707

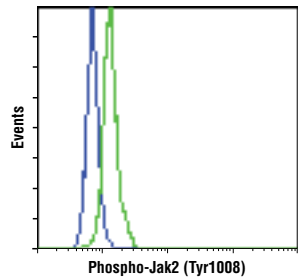
XP® Monoclonal Antibodies
one antibody, multiple applications™

Immunology and Inflammation

	Applications	Reactivity
#4327 Phospho-AML1 (Ser249) Antibody	W, IP, IF-IC, F	H
XP #4336 AML1 (D33G6) XP® Rabbit mAb	W, IHC-P, IF-IC, F	H, Mk
#4334 AML1 Antibody	W, IF-IC, F	H, Mk
New #8638 BATF (D7C5) Rabbit mAb	W, IP, IF-IC, F	H, M
#4443 CD3ε (CD3-12) Rat mAb	W, IP, F	H, M, (Pg)
#3563 CD4 (Edu-2) Mouse mAb	F	H
#3572 CD8 (RIV11) Mouse mAb	F	H
#3565 CD10 (CB-CALLA) Mouse mAb	F	H
#3574 CD19 Antibody	W, IP, IF-IC, F	H, M
#3568 CD31 (PECAM-1) (158-2B3) Mouse mAb	F	H
#3528 CD31 (PECAM-1) (89C2) Mouse mAb	W, IP, IHC-P, IF-IC, F	H
#3569 CD34 (IC0115) Mouse mAb	IHC-P, F	H
#3570 CD44 (156-3C11) Mouse mAb	CA W, IP, IHC-P, IF-IC, F	H
#4041 CD44 (156-3C11) Mouse mAb (Biotinylated)	W, F	H
#5640 CD44 (8E2) Mouse mAb	W, IP, IF-IC, F	H, M, R
#5173 Phospho-CD79A (Tyr182) Antibody	W, IP, IF-IC, F	H, (M, R)
#4841 Cox1 Antibody	W, IP, F	H, M, R, Mk
#2294 DRAK2 (33D7) Rabbit mAb	W, IP, IHC-P, F	M
#2593 Evi-1 (C50E12) Rabbit mAb	W, IP, IF-IC, F	H
XP #4589 GATA-1 (D24E4) XP® Rabbit mAb	W, IP, IF-IC, F	H
XP #3535 GATA-1 (D52H6) XP® Rabbit mAb	W, IP, IHC-P, IF-IC, F	H, M, R
New XP #5852 GATA-3 (D13C9) XP® Rabbit mAb	W, IF-IC, F	H, (Mk)
New XP #8714 Phospho-HS1 (Tyr397) (D12C1) XP® Rabbit mAb	W, IF-IC, F	H, (M, R)
XP #3892 HS1 (D5A9) XP® Rabbit mAb (Rodent Specific)	W, IP, IHC-P, F	M, R
XP #3890 HS1 (D83A8) XP® Rabbit mAb (Human Specific)	W, IP, IHC-P, IF-IC, F	H
#4503 HS1 Antibody (Human Specific)	W, IP, F	H
#4814 IκBα (L35A5) Mouse mAb (Amino-terminal Antigen)	CA W, IP, IHC-P, IF-IC, F	H, M, R, Mk, B, Pg
XP #3416 IKKε (D61F9) XP® Rabbit mAb	W, IP, IF-IC, F	M, R
New XP #8478 IRF-1 (D5E4) XP® Rabbit mAb	W, IP, IF-IC, F	H, M, R, (Mk)
#4964 IRF-4 Antibody	W, IP, IF-IC, F, ChIP	H
New #8082 Phospho-Jak2 (Tyr1008) (D4A8) Rabbit mAb	W, F	H, M, (R, Mk, X, B, Pg)
XP #3230 Jak2 (D2E12) XP® Rabbit mAb	W, IP, IHC-P, IF-IC, F	H, M, R, (Hm, Mk, C, X, B, Dg, Pg)
XP #4040 Jak2 (D2E12) XP® Rabbit mAb (Biotinylated)	W, F	H, M, R
#3329 MND4 (3C1) Rat mAb	CA W, IF-IC, F	H
#9166 LAT Antibody	W, IP, IHC-P, F	H, M, (R)
#2787 Lck (73A5) Rabbit mAb	W, IF-IC, F	H
#2088 LEDGF (C57G11) Rabbit mAb	W, IHC-P, IF-IC, F	H, M, R, (Mk)
#3576 CD56 (NCAM) (123C3) Mouse mAb	W, IHC-P, IF-IC, F	H
New XP #5861 NFAT1 (D43B1) XP® Rabbit mAb	W, IP, IHC-P, IF-IC, F	H, M
#3017 NF-κB2 p100/p52 (18D10) Rabbit mAb (Human Specific)	W, IHC-P, F	H, Mk
#3033 Phospho-NF-κB p65 (Ser536) (93H1) Rabbit mAb	CA W, IP, IF-IC, F	H, M, R, Hm, Mk, Pg, (Dg)
New XP #8242 NF-κB p65 (D14E12) XP® Rabbit mAb	W, IP, IHC-P, IF-IC, F, ChIP	H, M, R, Hm, Mk, Dg
#4764 NF-κB p65 (C22B4) Rabbit mAb	W, IHC-P, IF-IC, F	H, M, R, Mk, B, (Dg)
New #6956 NF-κB p65 (L8F6) Mouse mAb	W, IP, IHC-P, IF-IC, F, ChIP	H, M, R, Hm, Mk, Mi, B, Dg, Pg
New XP #8970 PAX5 (D19F8) XP® Rabbit mAb	W, IP, IF-IC, F	H, M, (X)
XP #3550 PIAS1 (D33A7) XP® Rabbit mAb	W, IF-IC, F	H, M, R, Mk
#9777 Pirin (1E8) Rat mAb	W, IP, F	H, M, R, Hm, Mk, B
#2258 PU.1 (9G7) Rabbit mAb	CA W, IP, IHC-P, IF-IC, F, ChIP	H, M, (Mk, Pg)
#2266 PU.1 Antibody	W, IP, IHC-P, IF-IC, F, ChIP	H, M, (Mk, Pg)



GATA-3 (D13C9) XP® Rabbit mAb #5852: Flow cytometric analysis of THP-1 cells (blue) and MCF7 cells (green) using #5852.



Phospho-Jak2 (Tyr1008) (D4A8) Rabbit mAb #8082: Flow cytometric analysis of BaF3 cells, untreated (blue) or treated with mIL-3 #8923 (green), using #8082.

Product References:

GATA-3 (D13C9) XP® Rabbit mAb #5852: Malu, D.T. et al. (2011) Interferon-regulatory factor 4 is essential for the developmental program of T helper 9 cells. *J. Immunol.* 186, 6271–6279.

IκBα (L35A5) Mouse mAb (Amino-terminal Antigen) (Alexa Fluor® 488 Conjugate) #5743: Katzman, S.D. et al. (2010) Duration of antigen receptor signaling determines T-cell tolerance or activation. *Proc. Natl. Acad. Sci. U.S.A.* 107, 18085–18090.

IRF-4 Antibody #4964: Staudt, V. et al. (2010) Interferon-regulatory factor 4 is essential for the developmental program of T helper 9 cells. *Immunity* 33, 192–202.

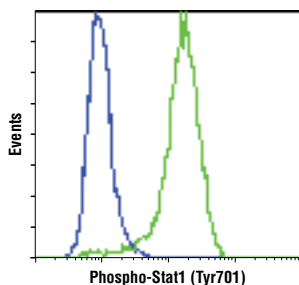
MND4 (3C1) Rat mAb #3329: McClintock-Treep, S.A. et al. (2011) Quantitative assessment of myeloid nuclear differentiation antigen distinguishes myelodysplastic syndrome from normal bone marrow. *Am. J. Clin. Pathol.* 135, 380–385.

Phospho-NF-κB p65 (Ser536) (93H1) Rabbit mAb #3033: Xie, S. et al. (2010) IL-17 activates the canonical NF-κappaB signaling pathway in autoimmune B cells of BXD2 mice to upregulate the expression of regulators of G-protein signaling 16. *J. Immunol.* 184, 2289–2296. / Dental, C. et al. (2011) Hepatitis C virus fails to activate NF-κB signaling in plasmacytoid dendritic cells. *J. Virol.* 86, 1090–1096.

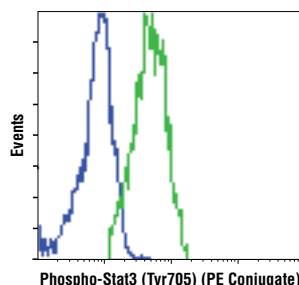
Phospho-NF-κB p65 (Ser536) (93H1) Rabbit mAb (Alexa Fluor® 647 Conjugate) #4887: Kalland, M.E. et al. (2011) T cell-signaling network analysis reveals distinct differences between CD28 and CD2 costimulation responses in various subsets and in the MAPK pathway between resting and activated regulatory T cells. *J. Immunol.* 187, 5233–5245.

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Immunology and Inflammation Continued



Phospho-Stat1 (Tyr701) (D4A7) Rabbit mAb #7649: Flow cytometric analysis of Jurkat cells, untreated (blue) or treated with hIFN- α 1 #8927 (green), using #7649.



Phospho-Stat3 (Tyr705) (D3A7) XP[®] Rabbit mAb (PE Conjugate) #8119: Flow cytometric analysis of Jurkat cells, untreated (blue) or treated with hIFN- α 1 #8927 (green), using #8119.

	Applications	Reactivity
#4727 c-Rel Antibody	W, IP, IHC-P, IF-IC, F	H, Mk
XP[®] #5025 Phospho-RelB (Ser552) (D41B9) XP [®] Rabbit mAb	W, IP, IF-IC, F	H, M, (R, Mk, B, Dg)
#4999 Phospho-RelB (Ser552) Antibody	W, IP, IF-IC, F	H, M, (R, Mk, B, Dg)
#2805 SH2D1A (XLP 1D12) Rat mAb	W, F	H
#9167 Phospho-Stat1 (Tyr701) (58D6) Rabbit mAb	CA W, IP, IHC-P, IHC-F, IF-IC, F, ChIP	H, M
New #7649 Phospho-Stat1 (Tyr701) (D4A7) Rabbit mAb	W, IP, IF-IC, F, ChIP	H, M, R, (Mk)
#9177 Phospho-Stat1 (Ser727) Antibody	W, IF-IC, F, ChIP	H, M, R, (B)
XP[®] #9145 Phospho-Stat3 (Tyr705) (D3A7) XP [®] Rabbit mAb	CA W, IP, IHC-P, IHC-F, IF-IC, F, ChIP	H, M, R, Mk, (Hm, B, Pg)
#9138 Phospho-Stat3 (Tyr705) (3E2) Mouse mAb	W, IP, F	H, M, (R, B)
#4113 Phospho-Stat3 (Tyr705) (M9C6) Mouse mAb	W, IP, IHC-P, IF-IC, F	H, M, R, Mk
#9139 Stat3 (124H6) Mouse mAb	W, IP, IHC-P, IF-IC, F, ChIP	H, M, R, Mk
New #4134 Phospho-Stat4 (Tyr693) (D2E4) Rabbit mAb	W, IP, IF-IC, F, ChIP	H, (M, R, Mk, Pg)
XP[®] #4322 Phospho-Stat5 (Tyr694) (D47E7) XP [®] Rabbit mAb	W, IP, IF-IC, F	H, M, (R, Mk, B)
#9359 Phospho-Stat5 (Tyr694) (C11C5) Rabbit mAb	W, IP, IHC-P, F	H, M, (R, Mk, B)
#9314 Phospho-Stat5 (Tyr694) (C71E5) Rabbit mAb	CA W, IHC-P, IF-IC, F	H, M, (R, Mk, B)
#9351 Phospho-Stat5 (Tyr694) Antibody	W, F, ChIP	H, M, (R, B)
#4807 Stat5a (4H1) Mouse mAb	W, IP, F	H
#9361 Phospho-Stat6 (Tyr641) Antibody	W, IP, IF-IC, F	H, (B)
XP[®] #5483 Phospho-TBK1/NAK (Ser172) (D52C2) XP [®] Rabbit mAb	W, IP, IF-IC, F	H, (M, R, Mk, X, B, Dg)
#5251 Phospho-TCTP (Ser46) Antibody	W, IHC-P, IF-IC, F	H, M, R, Mk
#2701 Phospho-Zap-70 (Tyr319)/Syk (Tyr352) Antibody	W, IP, IF-IC, F	H, M
#2704 Phospho-Zap-70 (Tyr493) Antibody	W, IP, F	H, (M, R)
XP[®] #3165 Zap-70 (D1C10E) XP [®] Rabbit mAb	CA W, IP, IF-F, F	H, M
#2705 Zap-70 (99F2) Rabbit mAb	W, IP, IHC-P, F	H, M

Product References:

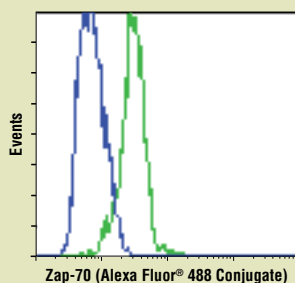
Phospho-Stat3 (Tyr705) (D3A7) XP[®] Rabbit mAb (Alexa Fluor[®] 488 Conjugate) #4323: Anand, S. et al. (2011) Increased basal intracellular signaling patterns do not correlate with JAK2 genotype in human myeloproliferative neoplasms. *Blood* 118, 1610–1621.

Zap-70

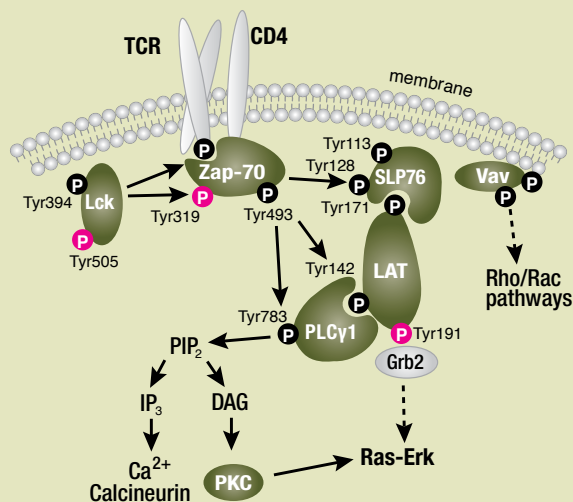
The Syk family protein tyrosine kinase Zap-70 is expressed in T and NK cells and plays a critical role in mediating T cell activation in response to T cell receptor (TCR) engagement. Following TCR engagement, Zap-70 is rapidly phosphorylated on several tyrosine residues through autophosphorylation and transphosphorylation by the Src family tyrosine kinase Lck. Tyrosine phosphorylation correlates with increased Zap-70 kinase activity and downstream signaling events. Researchers have found that expression of Zap-70 is correlated with disease progression and survival in patients with chronic lymphocytic leukemia.^{1,2}

¹ Wiestner, A. et al. (2003) *Blood* 101, 4944–4951.

² Crespo, M. et al. (2003) *N. Engl. J. Med.* 348, 1764–1775.

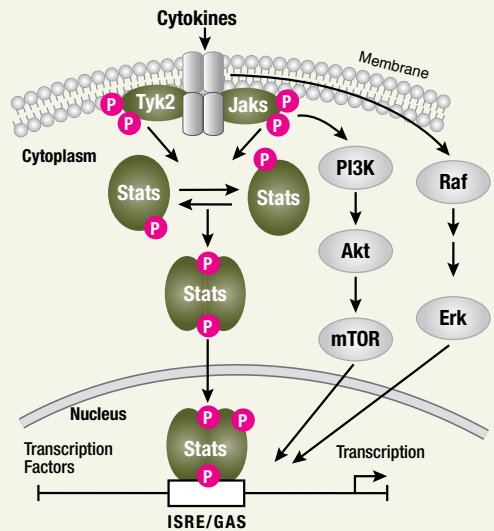


Zap-70 (D1C10E) XP[®] Rabbit mAb (Alexa Fluor[®] 488 Conjugate) #9473: Flow cytometric analysis of Ramos (blue) and Jurkat (green) cells using #9473.



Jak/Stat Signaling

Cytokine signaling is integral to an efficient immune response. A key pathway involved in cytokine signaling is the Janus kinase-signal transducer and activator of transcription (Jak/Stat) pathway. Jaks and Stats regulate growth, survival, differentiation, and pathogen resistance. Cytokine binding induces receptor dimerization, thereby activating the associated Jaks, which undergo autophosphorylation and subsequently phosphorylate the receptor. These phosphorylated sites serve as docking sites for the SH2 domain-containing Stats, such as Stat3, and for SH2-containing proteins and adaptors that link the receptor to MAP kinase, PI3K/Akt, and other cellular pathways. Receptor-bound Stats phosphorylated by Jaks dimerize and translocate to the nucleus where they regulate target gene transcription.



Jak/Stat Utilization Table

Combinatorial use of Tyrosine Kinases and Stat Proteins in Cytokine/Growth Factor Signaling

Ligand	Receptor	Jak-kinase	Other Tyrosine Kinases	Stat Family Members
IL-6	IL-6Ra+gp130	Jak1, Jak2, Tyk2	Hck	Stat1, Stat3
IL-11	IL-11R+gp130	Jak1, Jak2, Tyk2	Src, Yes	Stat3
CNTF, CT-1, LIF, OSM	CNTFR+gp130, CT-1R+gp130, LIFR+gp130, OSMR+gp130	Jak1, Jak2, Tyk2	Src family	Predominant: Stat3 Secondary: Stat1, Stat5
G-CSF	G-CSFR	Jak2, Tyk2	Lyn	Stat3
IL-12 (p40+p35)	IL-12Rβ1+IL-12Rβ2	Jak2, Tyk2	Lck	Stat4
Leptin	LeptinR	Jak2	not determined	Stat3, Stat5, Stat6
IL-3	IL-3Ra+βc	Jak2	Fyn, Hck, Lyn	Stat3, Stat5, Stat6
IL-5	IL-5R+βc	Jak2	Btk	Stat3, Stat5, Stat6
GM-CSF	GM-CSFR+βc	Jak2	Hck, Lyn	Stat3, Stat5
Angiotensin	GPCR	Jak2, Tyk2		Stat1, Stat2, Stat3
Serotonin	GPCR	Jak2		Stat3
α-Thrombin	GPCR	Jak2		Stat1, Stat3
Chemokines	CXCR4	Jak2, Jak3		
IL-2	IL-2Ra+IL-2Rb+yc	Jak1, Jak2, Jak3	Fyn, Hck, Lck, Syk, Tec	Stat3, Stat5
IL-4	IL-4Ra+yc or IL-4Ra+IL-13Ra1	Jak1, Jak3	Lck, Tec	Stat6
IL-7	IL-7R+yc	Jak1, Jak3	Lyn	Stat3, Stat5
IL-9	IL-9R+yc	Jak1, Jak3	not determined	Stat1, Stat3, Stat5
IL-13	IL-13Ra1+IL-4Ra	Jak1, Jak2, Tyk2	Ctk	Stat6
IL-15	IL-15Ra+IL-2Rβ+yc	Jak1, Jak3	Lck	Stat3, Stat5
IL-19	IL-20Ra+IL-20Rβ	Jak1, ?		Stat3
IL-20	IL-20Ra, IL-22R+IL-20Rβ	Jak1, ?		Stat3
IL-21	IL-21R+yc	Jak1, Jak3		Stat1, Stat3, Stat5
IL-22	IL-22R+IL-10Rβ	Jak1, Tyk2		Stat1, Stat3, Stat5
IL-23 (p40+p19)	IL-12Rβ1+IL-23R	Jak2, Tyk2		Stat4
IL-24	same as IL-20	Jak1, ?		Stat3
IL-26	IL-20Ra+IL-10Rβ	Jak1, Tyk2		Stat3
IL-27 (EBI3+p28)	gp130+WSX1	Jak1, Jak2, Tyk2		Stat1, Stat2, Stat3, Stat4, Stat5
IL-28A, IL-28B, IL-29	IL-28R+IL-10Rβ	Jak1, Tyk2		Stat1, Stat2, Stat3, Stat4, Stat5
IL-31	IL-31Ra+OSMR	Jak1, Jak2, Tyk2		Stat1, Stat3, Stat5
IL-35 (p35+EBI3)	gp130+WSX1	Jak1, Jak2, Tyk2		Stat1, Stat3, Stat5
GH	GHR	Jak2	Src family	Stat3, Stat5 (mainly Stat5a)
Tpo	TpoR (c-Mpl)	Tyk2, Jak2	Lyn	Stat1, Stat3, Stat5
Epo, Pro	EpoR, ProlactinR	Jak2	Src Family	Stat5 (mainly Stat5a)
Interferon (IFNα/β)	IFNAR1+IFNAR2	Jak1, Tyk2	Lck	Predominant: Stat1, Stat2 Secondary: Stat3, Stat4, Stat5
IFN-γ	IFN-γR1+IFN-γR2	Jak1, Jak2	Hck, Lyn	Stat1
IL-10	IL-10Ra+IL-10Rβ	Jak1, Tyk2	not determined	Stat3, Stat1, Stat5
TLSP	TLSPR and IL7R	Jak1, possibly Jak2	not determined	Stat3, Stat5
EGF	EGFR	Jak1	EGFR, Src	Stat1, Stat3, Stat5
PDGF	PDGFR	Jak1, Jak2	PDGFR, Src	Stat1, Stat3, Stat5

Bioactive Cytokines and Growth Factors

The world's highest quality research antibody provider has extended its expertise to Cytokine and Growth Factor production.

Quality

- Most are greater than 98% pure as demonstrated by SDS-PAGE.
- Endotoxin levels are tested by the LAL assay and are less than 0.01 ng/μg cytokine.
- Reduced and non-reduced protein is run on SDS-PAGE.
- ED₅₀ or maximum 50% response is determined by a standard cell based assay for every lot.
- Several lots are tested side-by-side to ensure consistent bioactivity.
- Bioactivity and purity data is shown on each product webpage and datasheet.

Consistency

- Strict specifications are set and enforced.
- Each lot is compared to previous lots for consistency in purity and bioactivity.
- Lyophilized lots are quality assured for sterility and bioactivity.

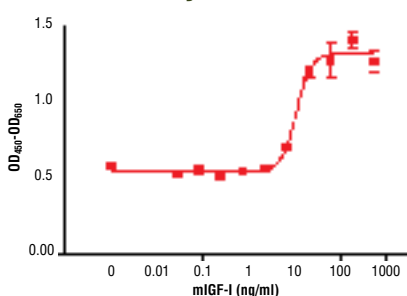
Dependability

- Products are produced in-house and ready to ship.
- Products are available in multi-milligram sizes.
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- CST sales and technical support ensure the highest quality customer service support.

New	#8445 Human C-C Motif Chemokine 3 (hCCL3/MIP-1-α)
	#3583 Human CD40 Ligand (hCD40L)
	#5717 Human Cystatin C (hCystatin C)
	#8916 Human Epidermal Growth Factor (hEGF)
New	#5331 Mouse Epidermal Growth Factor (mEGF)
	#5493 Human Epigen
New	#6980 Human Erythropoietin (hEPO)
	#5494 Human Epregrulin
	#5452 Human His6Fas Ligand/TNFSF6 (hHis6FasL)
New	#8910 Human Basic Fibroblast Growth Factor (hFGF basic/FGF2)
	#5234 Human FGF acidic (hFGF acidic)
	#5414 Mouse Basic Fibroblast Growth Factor (mFGF basic/FGF2)
	#8924 Human Fms-related Tyrosine Kinase 3 Ligand (hFLT3L)
	#8930 Human Granulocyte Colony Stimulating Factor (hG-CSF)
	#8922 Human Granulocyte Macrophage Colony Stimulating Factor (hGM-CSF)
	#5191 Mouse Granulocyte Macrophage Colony Stimulating Factor (mGM-CSF)
	#8927 Human Interferon-α1 (hIFN-α1)
	#8901 Human Interferon-γ (hIFN-γ)
New	#5222 Mouse Interferon-γ (mIFN-γ)
	#8917 Human Insulin-like Growth Factor I (hIGF-I)
New	#9897 Mouse Insulin-like Growth Factor I (mIGF-I)
New	#5238 Human Insulin-like Growth Factor II (hIGF-II)
	#5236 Human Interleukin-1α (hIL-1α)
	#5273 Mouse Interleukin-1α (mIL-1α)
	#8900 Human Interleukin-1β (hIL-1β)
	#5204 Mouse Interleukin-1β (mIL-1β)
	#8907 Human Interleukin-2 (hIL-2)
	#5454 Human Interleukin-2 (hIL-2) (mammalian derived)
	#5201 Mouse Interleukin-2 (mIL-2)
	#8918 Human Interleukin-3 (hIL-3)
	#8923 Mouse Interleukin-3 (mIL-3)
	#8919 Human Interleukin-4 (hIL-4)
	#5208 Mouse Interleukin-4 (mIL-4)
	#8904 Human Interleukin-6 (hIL-6)

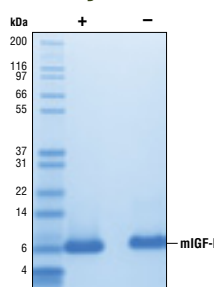
Mouse Insulin-like Growth Factor I (mIGF-I) #9897

Bioactivity



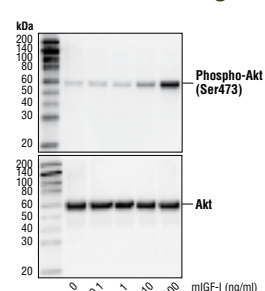
The ability of mIGF-I to induce phosphorylation of Akt was assessed. After serum starvation, NIH/3T3 cells were treated with increasing concentrations of mIGF-I for 10 min. Cells were lysed, and phospho-Akt was quantified using PathScan® Phospho-Akt (Thr308) Sandwich ELISA Kit #7252. OD₄₅₀ is shown.

Purity



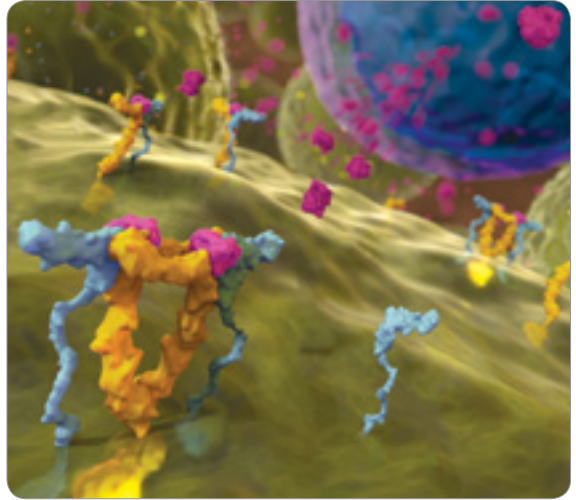
The purity of recombinant mIGF-I was determined by SDS-PAGE of 6 μg reduced (+) and non-reduced (-) recombinant mIGF-I and staining overnight with Coomassie Blue.

Downstream Signaling



Western blot analysis of extracts from NIH/3T3 cells, untreated or treated with increasing concentrations of mIGF-I for 10 min, using Phospho-Akt (Ser473) (D9E) XP® Rabbit mAb #4060 (upper) and Akt1 (C73H10) Rabbit mAb #2938 (lower).

- New** #5216 Mouse Interleukin-6 (mIL-6)
- New** #8170 Human Interleukin-7 (hIL-7)
- #5217 Mouse Interleukin-7 (mIL-7)
- New** #8921 Human Interleukin-8 (hIL-8)
- #8903 Human Interleukin-10 (hIL-10)
- #5358 Human Interleukin-10 (hIL-10) (mammalian derived)
- New** #5261 Mouse Interleukin-10 (mIL-10)
- #8905 Human Interleukin-13 (hIL-13)
- #5242 Mouse Interleukin-13 (mIL-13)
- #8928 Human Interleukin-17A (hIL-17A)
- #5227 Mouse Interleukin-17A (mIL-17A)
- New** #9584 Mouse Interleukin-17B (mIL-17B)
- #8906 Human Interleukin-17F (hIL-17F)
- #8920 Human Interleukin-21 (hIL-21)
- #8931 Human Interleukin-22 (hIL-22)
- #5224 Mouse Interleukin-22 (mIL-22)
- New** #5725 Mouse His₆Interleukin-27 (mHis₆IL-27)
- #5164 Human Interleukin-28A (hIL-28A/IFN- λ 2)
- New** #8796 Human Interleukin-28B (hIL-28B/IFN- λ 3)
- #5183 Human Interleukin-29 (hIL-29)
- New** #5719 Human Leptin/OB (hLeptin)
- #8929 Human Macrophage Colony Stimulating Factor (hM-CSF)
- New** #5228 Mouse Macrophage Colony Stimulating Factor (mM-CSF)
- #5221 Human β -Nerve Growth Factor (h β -NGF)
- #5218 Human Neuregulin-1 (hNRG-1)
- New** #5237 Human Neurotrophin-3 (hNT-3)
- New** #5592 Human Neurotrophin-4 (hNT-4)
- New** #5367 Human Oncostatin M (hOSM)
- #5371 Mouse Oncostatin M (mOSM)
- #8912 Human Platelet-Derived Growth Factor BB (hPDGF-BB)
- #8913 Human Platelet-Derived Growth Factor AA (hPDGF-AA)
- #8925 Human Stem Cell Factor (hSCF)
- New** #5223 Mouse Stem Cell Factor (mSCF)
- New** #5712 Human Stromal Cell-derived Factor 1 β /CXCL12 (hSDF1 β)

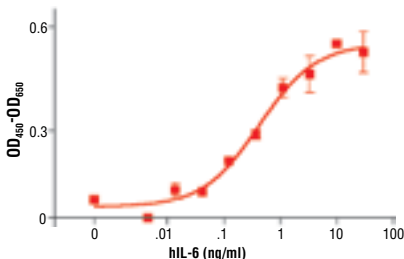


IL-6 (pink) is a potent inducer of the acute phase response and is produced by T cells, macrophages, fibroblasts, endothelial and other cells. IL-6 induces proliferation and differentiation, and in concert with TGF- β is important for developing Th17 responses. IL-6 binds to IL-6Ra (light blue) inducing gp130 (yellow) homodimerization. gp130 homodimerization triggers the Jak/STAT cascade and the SHP2/Erk MAP Kinase cascade. IL-6, through increasing expression of proangiogenic VEGF, may contribute to metastatic breast cancer.

- #5495 Human Transforming Growth Factor α (hTGF- α)
- #5154 Human Latent Transforming Growth Factor β 1 (hLatent TGF- β 1)
- #8915 Human Transforming Growth Factor β 1 (hTGF- β 1)
- #5231 Mouse Transforming Growth Factor β 1 (mTGF- β 1)
- #8406 Human Transforming Growth Factor β 2 (hTGF- β 2)
- #8425 Human Transforming Growth Factor β 3 (hTGF- β 3)
- #8902 Human Tumor Necrosis Factor- α (hTNF- α)
- #4698 Mouse His₆Tumor Necrosis Factor- α (mHis₆TNF- α)
- #5178 Mouse Tumor Necrosis Factor- α (mTNF- α)
- New** #8230 Human Lymphotoxin- α /TNF- β /TNFSF1 (hLT- α)
- New** #8460 Human His₆41BB Ligand/TNFSF9 (hHis₆4-1BBL)
- New** #5413 Human His₆BAFF/TNFSF13B (hHis₆ BAFF)
- New** #5233 Human BAFF/TNFSF13B (hBAFF)
- New** #8012 Human His₆Thymic Stromal Lymphopoietin (hHis₆TSLP)
- #5314 Mouse Vascular Endothelial Growth Factor-120 (mVEGF₁₂₀)
- #8908 Human Vascular Endothelial Growth Factor-121 (hVEGF₁₂₁)
- #5211 Mouse Vascular Endothelial Growth Factor-164 (mVEGF₁₆₄)
- New** #5874 Rat Vascular Endothelial Growth Factor-164 (rVEGF₁₆₄)
- #8065 Human Vascular Endothelial Growth Factor-165 (hVEGF₁₆₅)

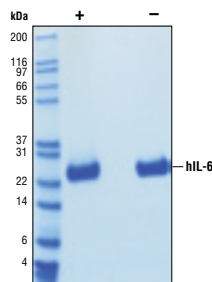
Human Interleukin-6 (hIL-6) #8904

Bioactivity



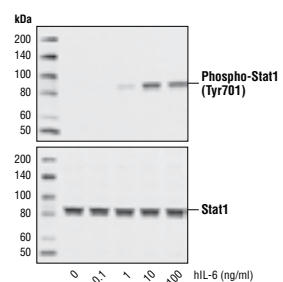
The proliferation of TF-1 cells treated with increasing concentrations of hIL-6 was assessed. After 48 hr treatment with hIL-6, cells were incubated with a tetrazolium salt and the OD₄₅₀ - OD₆₅₀ was determined.

Purity



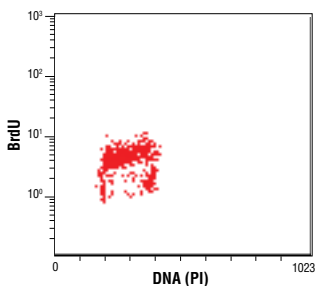
The purity of recombinant hIL-6 was determined by SDS-PAGE of 6 μ g reduced (+) and non-reduced (-) recombinant hIL-6 and staining overnight with Coomassie Blue.

Downstream Signaling

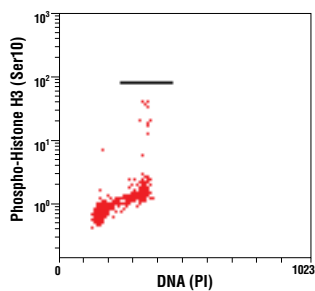


Western blot analysis of extracts from TF-1 cells, untreated or treated with hIL-6 for 10 min, using Phospho-Stat1 (Tyr701) Antibody #9171 (upper) and Stat1 Antibody #9172 (lower).

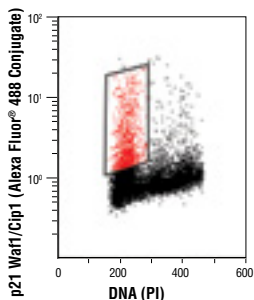
Cell Cycle, Checkpoint Control, and DNA Damage



BrdU (Bu20a) Mouse mAb #5292:
Flow cytometric analysis of Jurkat cells, incorporated with BrdU (30 min), using #5292 and Propidium Iodide (PI) #4087.



Phospho-Histone H3 (Ser10) (D2C8) XP® Rabbit mAb #3377: Flow cytometric analysis of Jurkat cells using #3377 and Propidium Iodide (PI) #4087. The gated population indicates Phospho-Histone H3 (Ser10)-positive cells.



p21 Waf1/Cip1 (12D1) Rabbit mAb (Alexa Fluor® 488 Conjugate) #5487:
Flow cytometric analysis of COS-7 cells using #5487 and Propidium Iodide (PI) #4087. Red = positive cells.

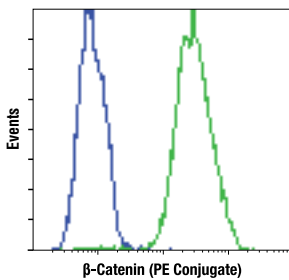
		Applications	Reactivity
#2675	Phospho-53BP1 (Ser1778) Antibody	W, IF-IC, F	H, Mk
XP	#2914 Phospho-Aurora A (Thr288)/Aurora B (Thr232)/Aurora C (Thr198) (D13A11) XP® Rabbit mAb	CA W, IF-IC, F	H, M, R
#3094	Aurora B/AIN1 Antibody	W, IP, F	H, M, R, Mk
#5292	BrdU (Bu20a) Mouse mAb	IHC-P, IF-IC, F	All
#4539	Phospho-cdc2 (Tyr15) (10A11) Rabbit mAb	W, IP, IF-IC, F	H, M, R, Mk
#9529	Phospho-cdc25C (Ser198) Antibody	W, F	H
#2561	Phospho-CDK2 (Thr160) Antibody	W, IP, F, E-P	H, M, R
#2546	CDK2 (78B2) Rabbit mAb	W, IP, F	H, M, R, Mk
#2316	CDK9 (C12F7) Rabbit mAb	W, IP, IHC-P, IHC-F, IF-IC, F	H, M, R, Hm, Mk, B, Dg
#2348	Phospho-Chk1 (Ser345) (133D3) Rabbit mAb	W, IF-IC, F	H, M, R, Mk
#2197	Phospho-Chk2 (Thr68) (C13C1) Rabbit mAb	W, IP, IHC-P, F	H, (Mk)
#2661	Phospho-Chk2 (Thr68) Antibody	W, IP, IF-IC, F	H, Mk
#4656	Cyclin A (BF683) Mouse mAb	W, F	H
#4135	Cyclin B1 (V152) Mouse mAb	CA W, F	H, M, (Hm)
XP	#3300 Phospho-Cyclin D1 (Thr286) (D29B3) XP® Rabbit mAb	W, IP, IF-IC, F	H, (Mk)
#2978	Cyclin D1 (92G2) Rabbit mAb	W, IHC-P, F	H, M, R
#4136	Phospho-Cyclin E (Thr62) Antibody	W, IP, IHC-P, F	H
#9718	Phospho-Histone H2A.X (Ser139) (20E3) Rabbit mAb	CA W, IHC-P, IF-IC, F	H, M, R, Mk
#2577	Phospho-Histone H2A.X (Ser139) Antibody	W, IHC-P, IF-IC, F	H, M, R
#5438	Phospho-Histone H2A.X (Ser139/Tyr142) Antibody	W, IP, IF-IC, F	H, M, R, Mk
XP	#3377 Phospho-Histone H3 (Ser10) (D2C8) XP® Rabbit mAb	CA W, IF-IC, F	H, M, R, Mk, Z
XP	#3642 Phospho-Histone H3 (Ser10) (D2C8) XP® Rabbit mAb (Biotinylated)	W, IF-F, IF-IC, F	H, M, R, Mk
#9701	Phospho-Histone H3 (Ser10) Antibody	CA W, IP, IHC-P, IHC-F, IF-IC, F	H, M, R, Mk, C, Dm, Z, Sc, (X)
#9706	Phospho-Histone H3 (Ser10) (6G3) Mouse mAb	W, IF-F, IF-IC, F	H, M, R
#9767	Phospho-Histone H3 (Thr11) (C2A6) Rabbit mAb	W, IP, F	H, M, R, (X)
#9764	Phospho-Histone H3 (Thr11) Antibody	W, IP, IF-IC, F	H, M, R, (X)
#9713	Phospho-Histone H3 (Ser28) Antibody	W, IP, IF-F, IF-IC, F	H, M, Hm, Dm, (R, C, X, Z, B)
#9649	Acetyl-Histone H3 (Lys9) (C5B11) Rabbit mAb	CA W, IHC-P, IF-IC, F, ChIP	H, M, R, Mk, Z, (Sc)
New XP	#5545 hnRNP A0 (D8A3) XP® Rabbit mAb	W, IP, IF-IC, F	H, M, R, Mk
New	#8443 hnRNP A1 (D21H11) Rabbit mAb	W, IP, IF-IC, F	H, M, R, Mk
#9304	hnRNP A2/B1 (2A2) Mouse mAb	W, F	H, Mk
#2616	HP1α Antibody	W, IP, IHC-P, IF-IC, F	H, M, R, Mk, (B)
#2619	HP1γ Antibody	W, IP, IF-IC, F	H, M, R, Mk
#4944	HR6A/HR6B Antibody	W, IF-IC, F	H, M, R, Mk, (C, Dm, X, Z)
#2786	INCENP (A841) Antibody	W, IF-IC, F	H
#2807	INCENP (P240) Antibody	W, IF-IC, F	H
#4822	p15 INK4B Antibody	W, F	H, M, R
#2753	Ku80 Antibody	W, IP, IHC-P, IF-IC, F	H, Mk, (M, R)
#2139	LSD1 Antibody	W, IP, IHC-P, IF-IC, F	H, M, R, Mk
#3515	MLH1 (4C9C7) Mouse mAb	W, IP, IF-IC, F	H, Mk
#4847	Mre11 (31H4) Rabbit mAb	W, IP, IHC-P, IHC-F, F	H
XP	#2017 MSH2 (D24B5) XP® Rabbit mAb	W, IP, IHC-P, IF-IC, F	H, M, Mk
XP	#3520 Phospho-NPM (Ser4) (D19C1) XP® Rabbit mAb	W, F	H, (M, R, Mk, X, B, Dg)
#3517	Phospho-NPM (Thr95) Antibody	W, IP, IF-IC, F	H, (M, R, Mk)
#2947	p21 Waf1/Cip1 (12D1) Rabbit mAb	CA W, IP, IHC-P, IF-IC, F	H, Mk
#9286	Phospho-p53 (Ser15) (16G8) Mouse mAb	CA W, IF-IC, F	H
#4030	Phospho-p53 (Ser15) (16G8) Mouse mAb (Biotinylated)	W, F	H
#9289	Phospho-p53 (Ser37) Antibody	W, IF-IC, F	H, Mk
#2521	Phospho-p53 (Ser46) Antibody	W, IP, IF-IC, F	H, Mk

Product References:

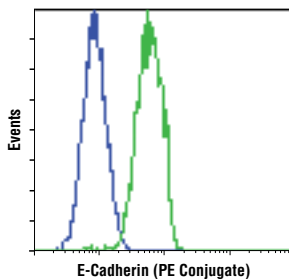
Phospho-Histone H3 (Ser10) (D2C8) XP® Rabbit mAb (Alexa Fluor® 488 Conjugate) #3465: Mathews, H.L. et al. (2011) Epigenetic patterns associated with the immune dysregulation that accompanies psychosocial distress. *Brain Behav. Immun.* 25, 830–839.

Phospho-Histone H3 (Ser10) Antibody (Alexa Fluor® 647 Conjugate) #9716: Kalland, M.E. et al. (2011) T cell-signaling network analysis reveals distinct differences between CD28 and CD2 costimulation responses in various subsets and in the MAPK pathway between resting and activated regulatory T cells. *J. Immunol.* 187, 5233–5245.

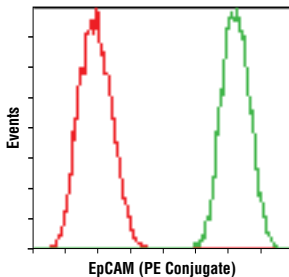
Stem Cell Markers, Development, and Differentiation



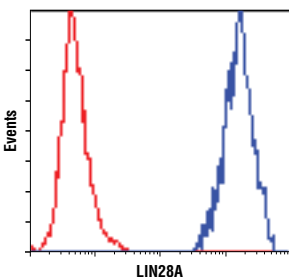
β-Catenin (L54E2) Mouse mAb (PE Conjugate) #6898: Flow cytometric analysis of NCI-H28 (blue) and HeLa (green) cells using #6898.



E-Cadherin (24E10) Rabbit mAb (PE Conjugate) #7559: Flow cytometric analysis of HeLa (blue) and MCF7 (green) cells using #7559.



EpCAM (VU1D9) Mouse mAb (PE Conjugate) #8995: Flow cytometric analysis of unpermeabilized HT-29 cells using #8995 (green) compared to concentration-matched Mouse (G3A1) mAb IgG1 Isotype Control (PE Conjugate) #6899 (red).



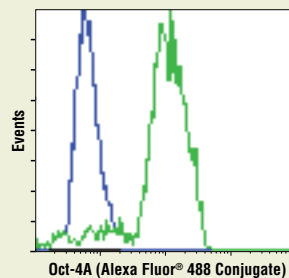
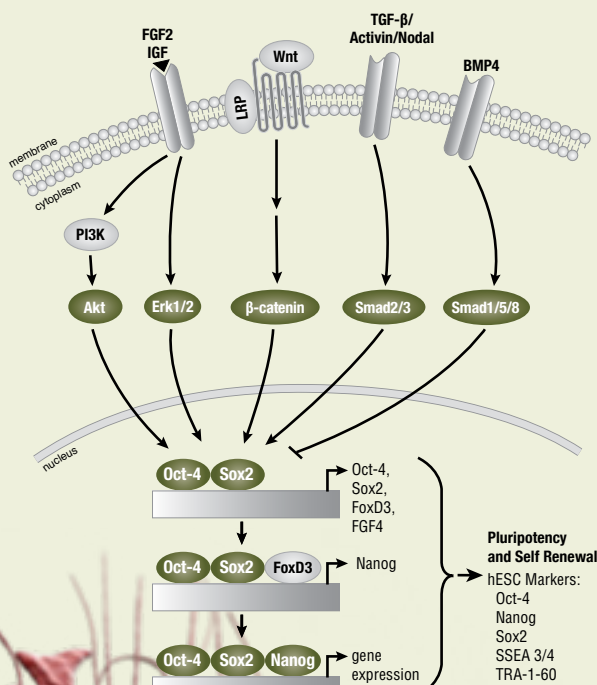
LIN28A (D1A1A) XP® Rabbit mAb #8641: Flow cytometric analysis of HeLa (red) and NTERA-2 (blue) cells using #8641.

		Applications	Reactivity
New	#8225 CACYBP (D43G11) Rabbit mAb	W, IHC-P, F	H, M, R, Mk
	#3354 CACYBP Antibody	W, IHC-P, F	H, M, R, Mk
New XP	#8480 β-Catenin (D10A8) XP® Rabbit mAb	W, IP, IHC-P, IHC-F, IF-F, IF-IC, F, ChIP	H, M, R, Mk, (Z, B, Pg)
	#3195 E-Cadherin (24E10) Rabbit mAb	CA W, IHC-P, IHC-F, IF-IC, F	H, M, (Dg, Pg)
	#2677 β-Catenin (L54E2) Mouse mAb (IF Preferred)	CA IP, IF-IC, F	H, (M, R, Mk, Pg)
	#5265 β-Catenin (L54E2) Mouse mAb (Biotinylated)	CA F	H
	#2929 EpCAM (VU1D9) Mouse mAb	CA W, IHC-P, IF-IC, F	H
XP	#4402 FoxP1 (D35D10) XP® Rabbit mAb	W, IP, IHC-P, F	H, M, R, Mk
	#2005 FoxP1 Antibody	W, IP, IHC-P, IF-IC, F	H, M, (R)
	#2230 LEF1 (C12A5) Rabbit mAb	CA W, IP, IF-IC, F	H, M, R
	#2286 LEF1 (C18A7) Rabbit mAb	W, IP, F	H, M
New XP	#8641 LIN28A (D1A1A) XP® Rabbit mAb	W, IF-IC, F	H, M, (R, Mk)
XP	#3695 LIN28A (D84C11) XP® Rabbit mAb	W, IF-IC, F	H, (R, Mk)
	#3978 LIN28A (A177) Antibody	W, IP, IHC-P, IF-IC, F	H, M, (Mk)
XP	#4903 Nanog (D73G4) XP® Rabbit mAb	CA W, IHC-P, IF-IC, F	H, (Mk)
	#3580 Nanog Antibody	W, IF-IC, F, ChIP	H
	#4893 Nanog (1E6C4) Mouse mAb	W, IHC-P, IF-IC, F	H
XP	#4380 Notch1 (D6F11) XP® Rabbit mAb	W, IF-IC, F	H, M, R
XP	#5732 Notch2 (D76A6) XP® Rabbit mAb	W, IP, IF-IC, F	H, M, R
	#2756 Numb (C29G11) Rabbit mAb	W, IP, IF-IC, F	H, M, R, Mk
	#2761 Numb (C44B4) Rabbit mAb	W, IP, IF-IC, F	H
	#2840 Oct-4A (C30A3) Rabbit mAb	CA W, IF-IC, F	H, M
	#2890 Oct-4A (C52G3) Rabbit mAb	W, IHC-P, IF-IC, F, ChIP	H
	#2750 Oct-4 Antibody	W, IHC-P, IF-IC, F, ChIP	H, (Mk)
	#9516 Phospho-Smad1/5 (Ser463/465) (41D10) Rabbit mAb	W, IF-IC, F	H, M, R
New XP	#6944 Smad1 (D59D7) XP® Rabbit mAb	W, IP, IF-IC, F, ChIP	H, M, (Mk, X, B)
XP	#9510 Phospho-Smad2 (Ser465/467)/ Smad3 (Ser423/425) (D6G10) XP® Rabbit mAb	IF-IC, F	H
XP	#5339 Smad2 (D43B4) XP® Rabbit mAb	W, IP, IF-IC, F, ChIP	H, M, R, Mk
New XP	#8685 Smad2/3 (D7G7) XP® Rabbit mAb	W, IP, IF-IC, F, ChIP	H, M, R, Mk
	#5678 Smad2/3 Antibody	W, IP, IF-IC, F, ChIP	H, M, R, Mk, (X)
	#9523 Smad3 (C67H9) Rabbit mAb	W, IP, IF-IC, F, ChIP	H, M, R, Mk, (X, Z, B)
XP	#3579 Sox2 (D6D9) XP® Rabbit mAb	CA W, IHC-P, IF-IC, F	H, (Mk, B, Dg)
	#4900 Sox2 (L1D6A2) Mouse mAb	W, IF-IC, F	H, M, (R, B, Dg)
	#4744 SSEA1 (MC480) Mouse mAb	CA IHC-P, IF-IC, F	M
	#2203 TCF1 (C63D9) Rabbit mAb	W, IP, IHC-P, IF-IC, F	H, M
	#4746 TRA-1-60(S) (TRA-1-60(S)) Mouse mAb	W, IHC-P, IF-IC, F	H
	#4745 TRA-1-81 (TRA-1-81) Mouse mAb	IHC-P, IF-IC, F	H
XP	#5741 Vimentin (D21H3) XP® Rabbit mAb	CA W, IHC-P, IF-IC, F	H, M, R, Mk

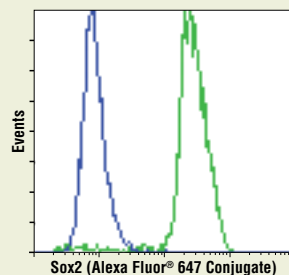
XP® Monoclonal Antibodies
one antibody, multiple applications™

Pluripotency in Human Embryonic Stem Cells

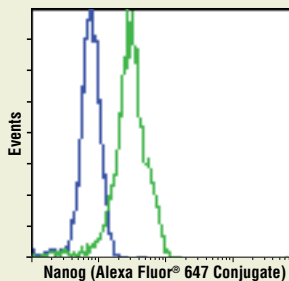
The predominant signaling pathways involved in human embryonic stem cell (ESC) pluripotency and self renewal are TGF- β , which signals through Smad2/3, and FGFR, which activates the MAPK and Akt pathways. The Wnt pathway also promotes pluripotency, although this may occur through a non-canonical mechanism involving a balance between the transcriptional activator TCF1 and the repressor TCF3. Signaling through these pathways results in the expression and activation of three key transcription factors: Oct-4, Sox2, and Nanog. These transcription factors activate gene expression of ESC-specific genes, regulate their own expression, and also serve as human ESC markers.



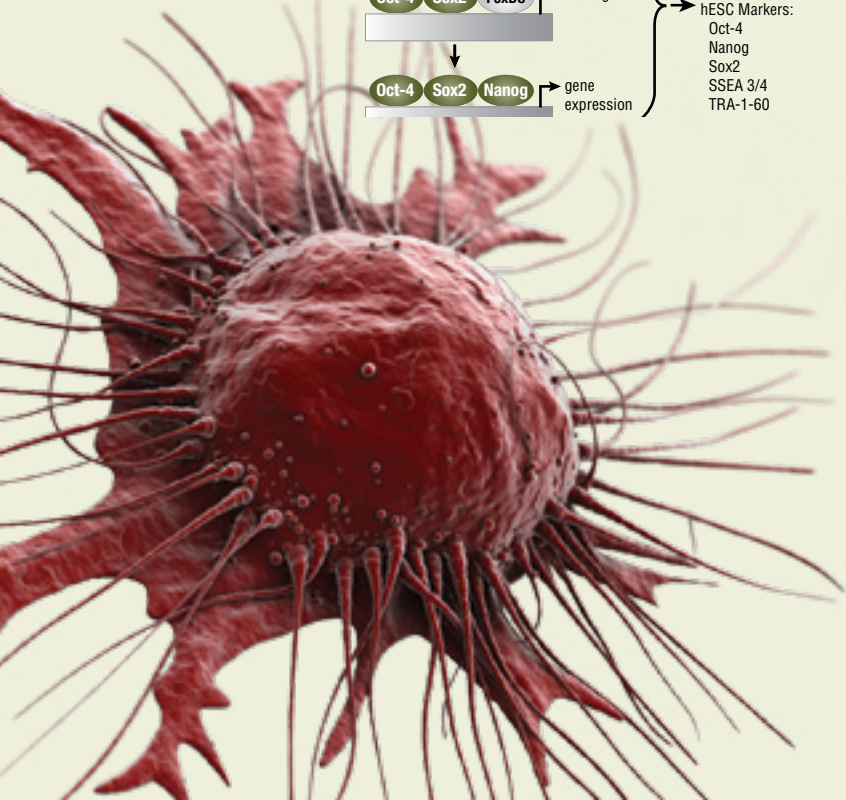
Oct-4A (C30A3) Rabbit mAb (Alexa Fluor® 488 Conjugate) #5177: Flow cytometric analysis of HeLa (blue) and NTERA-2 (green) cells using #5177.



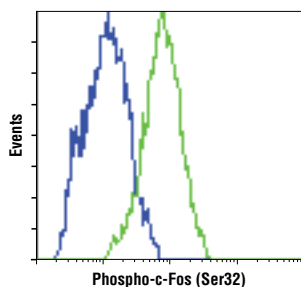
Sox2 (D6D9) XP® Rabbit mAb (Alexa Fluor® 647 Conjugate) #5067: Flow cytometric analysis of HeLa (blue) and NTERA-2 (green) cells using #5067.



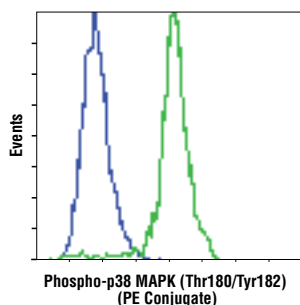
Nanog (D73G4) XP® Rabbit mAb (Alexa Fluor® 647 Conjugate) #5448: Flow cytometric analysis of HeLa (blue) and NTERA-2 (green) cells using #5448.



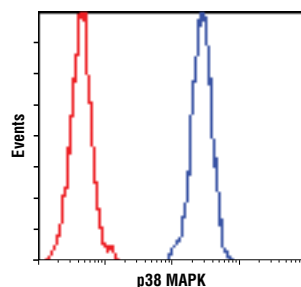
MAP Kinase Signaling



Phospho-c-Fos (Ser32) (D82C12) XP® Rabbit mAb #5348: Flow cytometric analysis of HeLa cells, untreated (blue) or treated with TPA #4174 (green), using #5348.



Phospho-p38 MAPK (Thr180/Tyr182) (3D7) Rabbit mAb (PE Conjugate) #6908: Flow cytometric analysis of THP-1 cells, untreated (blue) or anisomycin-treated (green), using #6908.



p38 MAPK (D13E1) XP® Rabbit mAb #8690: Flow cytometric analysis of HeLa cells using #8690 (blue) compared to concentration-matched Rabbit (DA1E) mAb IgG XP® Isotype Control #3900 (red).

Product References:

Phospho-p38 MAPK (Thr180/Tyr182) (28B10) Mouse mAb (Alexa Fluor® 647 Conjugate) #4552: Kalland, M.E. et al. (2011) T cell-signaling network analysis reveals distinct differences between CD28 and CD2 costimulation responses in various subsets and in the MAPK pathway between resting and activated regulatory T cells. *J. Immunol.* 187, 5233–5245.

Phospho-p44/42 MAPK (Erk1/2) (Thr202/Tyr204) (D13.14.4E) XP® Rabbit mAb #4370: Koenigsman, J. et al. (2009) N1 haploinsufficiency and Icsbp deficiency synergize in the development of leukemias. *Blood* 113, 4690–4701.

Phospho-p44/42 MAPK (Erk1/2) (Thr202/Tyr204) (197G2) Rabbit mAb #4377: Gross, A.J. et al. (2009) Developmental acquisition of the Lyn-CD22-SHP-1 inhibitory pathway promotes B cell tolerance. *J. Immunol.* 182, 5382–5392. / Messal, N. et al. (2011) Differential role for CD277 as a co-regulator of the immune signal in T and NK cells. *Eur. J. Immunol.* 41, 3443–3454.

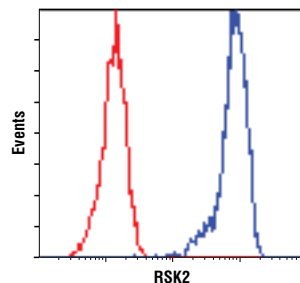
	Applications	Reactivity
#5112 Phospho-ATF-2 (Thr71) (11G2) Rabbit mAb	W, F	H, M, R, Mk
#9221 Phospho-ATF-2 (Thr71) Antibody	W, IP, IHC-P, IHC-F, IF-IC, F	H, M, R, Mk
#4067 Erk3 Antibody	W, IP, F	H, M, R, Mk
#5122 FAM129B Antibody	W, IP, IF-IC, F	H, Mk
XP #5348 Phospho-c-Fos (Ser32) (D82C12) XP® Rabbit mAb	W, IP, IF-IC, F	H, M, R, (Hm, Mk, B, Pg)
#2250 c-Fos (9F6) Rabbit mAb	W, IF-IC, F	H, M, R, (Hm, B, Pg)
#2251 FosB (5G4) Rabbit mAb	W, IP, IHC-P, IF-IC, F	H, M, R
#2023 FosB (5G4) Rabbit mAb (Alexa Fluor® 488 Conjugate)	F	H, M, Mk
New XP #5519 JIP4/SPAG9 (D72F4) XP® Rabbit mAb	W, IP, IHC-P, IF-IC, F	H, M, R, Mk
#9261 Phospho-c-Jun (Ser63) II Antibody	W, IP, IF-IC, F	H, M, R, Mk, Pg
XP #3270 Phospho-c-Jun (Ser73) (D47G9) XP® Rabbit mAb	W, IP, IHC-P, IF-IC, F	H, M, R, Mk
New #5000 JunD (D17G2) Rabbit mAb	W, IP, IF-IC, F	H, Mk, B, Pg
#3007 Phospho-MAPKAPK-2 (Thr334) (27B7) Rabbit mAb	CA W, IHC-P, IF-IC, F	H, M, R, Mk
#3042 MAPKAPK-2 Antibody	W, IP, F	H, M, R, Mk, (Hm)
#4660 Phospho-MEK1/2 (Ser217/221) (41G9) Rabbit mAb (Alexa Fluor® 488 Conjugate)	F	H, M, R, Mk, Dm, (C)
#5068 Phospho-MEK1/2 (Ser217/221) (41G9) Rabbit mAb (Alexa Fluor® 647 Conjugate)	F	H, M, R, Mk, Dm, (C)
#2338 Phospho-MEK1/2 (Ser221) (166F8) Rabbit mAb	W, IHC-P, F	H, M, R, Mk, (Dg)
#9127 Phospho-MEK1 (Thr286) Antibody	W, IP, IF-IC, F	H, R, Mk, (M)
New #8727 MEK1/2 (D1A5) Rabbit mAb	W, IF-IC, F	H, M, R, Mk, (Hm, Dm, X, Z, B, Dg, Pg, Ce)
#4694 MEK1/2 (L38C12) Mouse mAb	W, IHC-P, IF-IC, F	H, M, R, Mk
XP #4511 Phospho-p38 MAPK (Thr180/Tyr182) (D3F9) XP® Rabbit mAb	W, IP, IHC-P, IF-IC, F	H, M, R, Mk, Sc, (Hm, C, Z, B, Pg)
#9215 Phospho-p38 MAPK (Thr180/Tyr182) (3D7) Rabbit mAb	CA W, IF-IC, F	H, M, R, Mk, Dm, Pg, Sc, (Hm, Z, B)
#4092 Phospho-p38 MAPK (Thr180/Tyr182) (3D7) Rabbit mAb (Biotinylated)	W, F	H, M, R, Mk, Dm, Pg, Sc
#9211 Phospho-p38 MAPK (Thr180/Tyr182) Antibody	W, IP, IF-IC, F	H, M, R, Mk, Dm, Pg, Sc, (Hm, Z, B)
#9216 Phospho-p38 MAPK (Thr180/Tyr182) (28B10) Mouse mAb	CA W, IP, IF-IC, F	H, M, R, Mk, Sc, (Z)
New XP #8690 p38 MAPK (D13E1) XP® Rabbit mAb	W, IHC-P, IF-IC, F	H, M, R, Hm, Mk, Pg
#9212 p38 MAPK Antibody	W, IHC-P, IF-IC, F	H, M, R, Mk, (C)
#9228 p38α MAPK (L53F8) Mouse mAb	W, IF-IC, F	H, M, R, Mk, Sc
XP #4370 Phospho-p44/42 MAPK (Erk1/2) (Thr202/Tyr204) (D13.14.4E) XP® Rabbit mAb	CA W, IP, IHC-P, IF-IC, F	H, M, R, Hm, Mk, Mi, Dm, Z, B, Dg, Pg, Sc, (Ce)
XP #4094 Phospho-p44/42 MAPK (Erk1/2) (Thr202/Tyr204) (D13.14.4E) XP® Rabbit mAb (Biotinylated)	W, F	H, M, R, Hm, Mk, Mi, Dm, Z, B, Pg, Sc
#4377 Phospho-p44/42 MAPK (Erk1/2) (Thr202/Tyr204) (197G2) Rabbit mAb	W, IF-IC, F	H, M, R, Mk, Mi, Dm, Z, Pg
#9101 Phospho-p44/42 MAPK (Erk1/2) (Thr202/Tyr204) Antibody	W, IP, IF-IC, F	H, M, R, Hm, Mk, Mi, Dm, Z, B, Pg, Ce, (C)
#9106 Phospho-p44/42 MAPK (Erk1/2) (Thr202/Tyr204) (E10) Mouse mAb	CA W, IP, F	H, M, R, Hm, Mk, Mi, Dm, Z, B, Pg
#4695 p44/42 MAPK (Erk1/2) (137F5) Rabbit mAb	CA W, IP, IHC-P, IF-IC, F	H, M, R, Hm, Mk, Mi, Dm, Z, B, Dg, Pg, Ce, (C)

Phospho-p44/42 MAPK (Erk1/2) (Thr202/Tyr204) (E10) Mouse mAb (Alexa Fluor® 647 Conjugate) #4375: Soond, D.R. et al. (2010) PI3K p110delta regulates T-cell cytokine production during primary and secondary immune responses in mice and humans. *Blood* 115, 2203–2213. / Kalland, M.E. et al. (2011) T cell-signaling network analysis reveals distinct differences between CD28 and CD2 costimulation responses in various subsets and in the MAPK pathway between resting and activated regulatory T cells. *J. Immunol.* 187, 5233–5245.

Phospho-MAPKAPK-2 (Thr334) (27B7) Rabbit mAb (Alexa Fluor® 647 Conjugate) #4320: Kalland, M.E. et al. (2011) T cell-signaling network analysis reveals distinct differences between CD28 and CD2 costimulation responses in various subsets and in the MAPK pathway between resting and activated regulatory T cells. *J. Immunol.* 187, 5233–5245.

New
XP

	Applications	Reactivity
#5013 p44/42 MAPK (Erk1/2) (137F5) Rabbit mAb (Biotinylated)	W, IP, F	H, M, R, Mk, Mi, Dm, Z, B, Pg
#9102 p44/42 MAPK (Erk1/2) Antibody	W, IP, IHC-P, IF-IC, F	H, M, R, Hm, Mk, Mi, Z, B, Pg, Sc
#4696 p44/42 MAPK (Erk1/2) (L34F12) Mouse mAb	W, IHC-P, IF-IC, F	H, M, R, Mk, Mi, Z, Pg
#9434 B-Raf (L12G7) Mouse mAb	W, IP, F	H, M, R, Mk, Dm
#5528 RSK2 (D21B2) XP [®] Rabbit mAb	W, IF-IC, F	H, M, R, Mk, B, Pg, (Dg)
#9255 Phospho-SAPK/JNK (Thr183/Tyr185) (G9) Mouse mAb	W, IP, IF-IC, F CA	H, M, R, Hm, Sc
#4514 Phospho-SEK1/MKK4 (Ser257) (C36C11) Rabbit mAb	W, F	H, M, R, Mk
#3225 TAB1 Antibody	W, IF-IC, F	H, M, R, Mk
#4505 TAK1 Antibody	W, F	H, M, R, Mk, B, (C, X)



RSK2 (D21B2) XP[®] Rabbit mAb #5528: Flow cytometric analysis of HeLa cells using #5528 (blue) compared to concentration-matched Rabbit (DA1E) mAb IgG XP[®] Isotype Control #3900 (red).

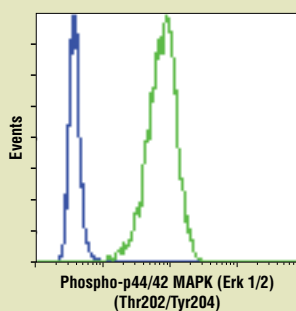
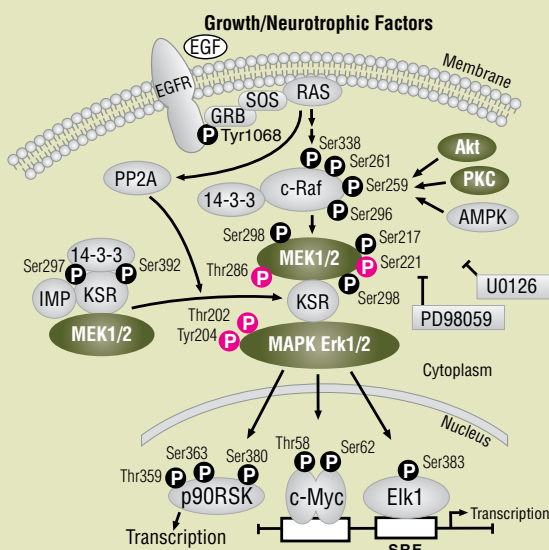
p44/42 MAPK

Mitogen-activated protein kinases (MAPKs) are a widely conserved family of serine/threonine protein kinases involved in many cellular programs such as cell proliferation, differentiation, motility, and death.

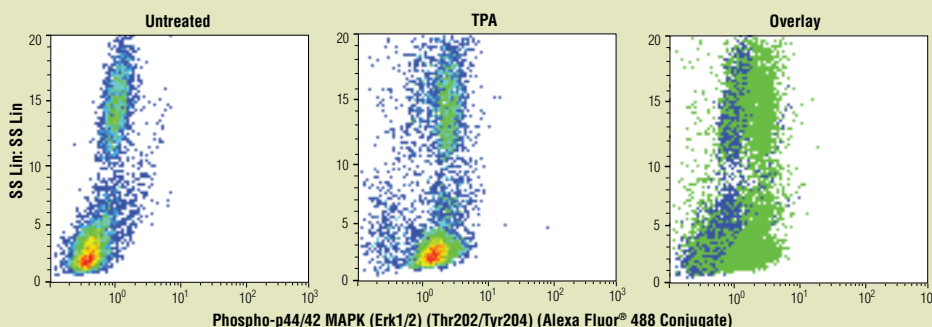
The p44/42 MAPK (Erk1/2) signaling pathway can be activated in response to a diverse range of extracellular stimuli including mitogens, growth factors, and cytokines and is an important target in the diagnosis and treatment of cancer.

Upon stimulation, a sequential three-part protein kinase cascade is initiated, consisting of a MAP kinase kinase kinase (MAPKKK or MAP3K), a MAP kinase kinase (MAPKK or MAP2K), and a MAP kinase (MAPK). Multiple p44/42 MAP3Ks have been identified, including members of the Raf family, as well as Mos and Tpl2/Cot. MEK1 and MEK2 are the primary MAPKKs in this pathway. MEK1 and MEK2 activate p44 and p42 through phosphorylation of activation loop residues Thr202/Tyr204 and Thr185/Tyr187, respectively.

Several downstream targets of p44/42 have been identified, including p90RSK and the transcription factor Elk-1. p44/42 are negatively regulated by a family of dual-specificity (Thr/Tyr) MAPK phosphatases, known as DUSPs or MKPs, along with MEK inhibitors such as U0126 and PD98059.

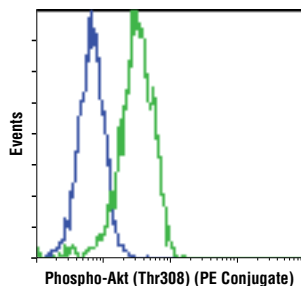


Phospho-p44/42 MAPK (Erk1/2) (Thr202/Tyr204) (D13.14.4E) XP[®] Rabbit mAb #4370: Flow cytometric analysis of Jurkat cells, treated with U0126 #9951 (blue) or TPA #4174 (green), using #4370.

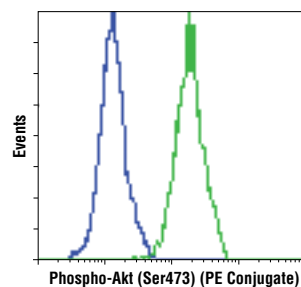


Phospho-p44/42 MAPK (Erk1/2) (Thr202/Tyr204) (E10) Mouse mAb (Alexa Fluor[®] 488 Conjugate) #4374: Flow cytometric analysis of whole blood, untreated or treated with TPA #4174, using #4374.

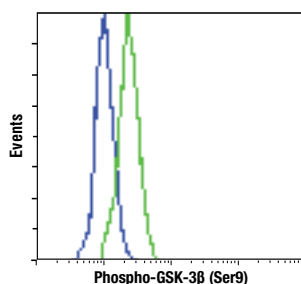
PI3K/Akt Signaling



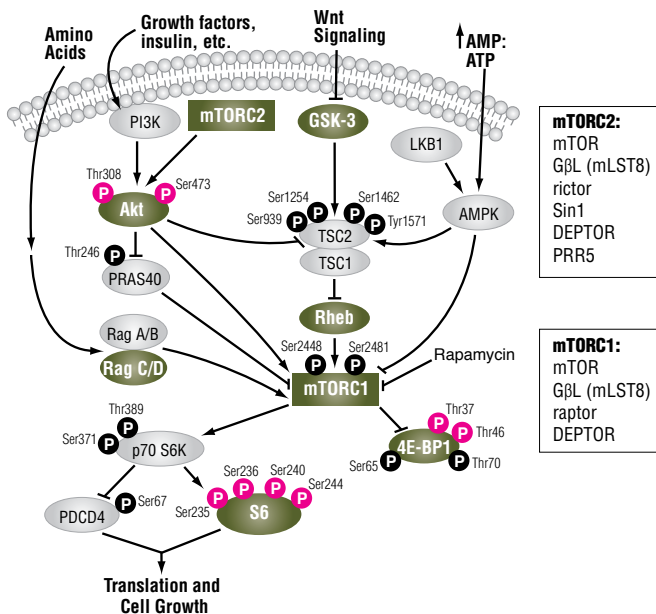
Phospho-Akt (Thr308) (C31E5E) Rabbit mAb (PE Conjugate) #9088: Flow cytometric analysis of NIH/3T3 cells, treated with hPDGF-BB #8912 (green) or with LY294002 #9901, Wortmannin #9951, and U0126 #9903 (blue), using #9088.



Phospho-Akt (Ser473) (D9E) XP® Rabbit mAb (PE Conjugate) #5315: Flow cytometric analysis of Jurkat cells, untreated (green) or treated with LY294002 #9901, Wortmannin #9551, and U0126 #9903 (blue), using #5315.



Phospho-GSK-3β (Ser9) (D85E12) XP® Rabbit mAb #5558: Flow cytometric analysis of NIH/3T3 cells, untreated (blue) or treated with hPDGF-BB #8912 (green), using #5558.



mTORC2:
mTOR
GβL (mLST8)
rictor
Sin1
DEPTOR
PRR5

mTORC1:
mTOR
GβL (mLST8)
raptor
DEPTOR

		Applications	Reactivity	
#2965	Phospho-Akt (Thr308) (C31E5E) Rabbit mAb	CA	W, IF-IC, F	H, M, R, Hm, Mk
#9275	Phospho-Akt (Thr308) Antibody		W, IP, F	H, M, R, Hm
XP®	#4060 Phospho-Akt (Ser473) (D9E) XP® Rabbit mAb	CA	W, IP, IHC-P, IHC-F, IF-IC, F	H, M, R, Hm, Mk, Dm, Z, B, (C, X, Dg, Pg)
XP®	#5012 Phospho-Akt (Ser473) (D9E) XP® Rabbit mAb (Biotinylated)		W, F	H, M, R, Hm, Dm, Z, B
#4058	Phospho-Akt (Ser473) (193H12) Rabbit mAb	CA	W, IP, IF-IC, F	H, M, R
#9271	Phospho-Akt (Ser473) Antibody		W, IP, IF-IC, F	H, M, R, Hm, Dm, B, Dg, Pg, (Mk, C, X)
#9272	Akt Antibody		W, IP, IF-IC, F	H, M, R, Hm, Mk, C, Dm, B, Pg, (Dg)
#2966	Akt (5G3) Mouse mAb	CA	IP, IF-IC, F	H, M, R, Hm
#4685	Akt (pan) (11E7) Rabbit mAb		W, IP, IHC-P, IF-IC, F	H, M, R, Mk
#4691	Akt (pan) (C67E7) Rabbit mAb	CA	W, IP, IHC-P, IF-IC, F	H, M, R, Mk, Dm
#2920	Akt (pan) (40D4) Mouse mAb		W, IP, IHC-P, IF-IC, F	H, M, R, Mk
#4821	Akt (pan) (40D4) Mouse mAb (Biotinylated)		W, IP, F	H, M, R, Mk
#4612	CTMP Antibody		W, IP, F	H, M, R
#2134	DJ-1 Antibody		W, F	H, M, R
XP®	#4818 GSK-3α (D80D1) XP® Rabbit mAb		IF-IC, F	H, M, (R)
XP®	#5558 Phospho-GSK-3β (Ser9) (D85E12) XP® Rabbit mAb		W, IP, IF-IC, F	H, M, R, Hm
New	#9832 GSK-3β (3D10) Mouse mAb		W, IP, IF-IC, F	H, M, R, Hm, Mk
XP®	#5482 Phospho-NDRG1 (Thr346) (D98G11) XP® Rabbit mAb	CA	W, IHC-P, IF-IC, F	H, M, R, Mk
#4912	YAP Antibody		W, IP, IHC-P, IF-IC, F	H, M, R, Mk

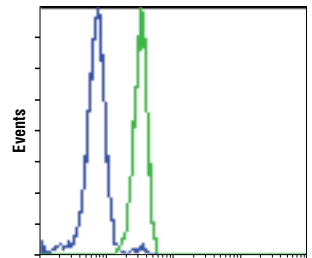
Product References:

- Phospho-Akt (Thr308) (C31E5E) Rabbit mAb #2965:** Sykes, S.M. et al. (2011) AKT/FOXO signaling enforces reversible differentiation blockade in myeloid leukemias. *Cell* 146, 697–708.
- Phospho-Akt (Ser473) (D9E) XP® Rabbit mAb #4060:** Sykes, S.M. et al. (2011) AKT/FOXO signaling enforces reversible differentiation blockade in myeloid leukemias. *Cell* 146, 697–708.
- Phospho-Akt (Ser473) (D9E) XP® Rabbit mAb (Alexa Fluor® 647 Conjugate) #4075:** Soond, D.R. et al. (2010) PI3K p110delta regulates T-cell cytokine production during primary and secondary immune responses in mice and humans. *Blood* 115, 2203–2213. / Kalland, M.E. et al. (2011) T cell-signaling network analysis reveals distinct differences between CD28 and CD2 costimulation responses in various subsets and in the MAPK pathway between resting and activated regulatory T cells. *J. Immunol.* 187, 5233–5245.
- Phospho-Akt (Ser473) (193H12) Rabbit mAb #4058:** Firaguy, G. and Nunès, J.A. (2009) Analysis of signaling events by dynamic phosphoflow cytometry. *Sci Signal* 2, pi3. / Messal, N. et al. (2011) Differential role for CD277 as a co-regulator of the immune signal in T and NK cells. *Eur. J. Immunol.* 41, 3443–3454.
- Phospho-Akt (Ser473) (193H12) Rabbit mAb (Alexa Fluor® 488 Conjugate) #2336:** Anand, S. et al. (2011) Increased basal intracellular signaling patterns do not correlate with JAK2 genotype in human myeloproliferative neoplasms. *Blood* 118, 1610–1621.
- Phospho-Akt (Ser473) (193H12) Rabbit mAb (Alexa Fluor® 647 Conjugate) #2337:** Mahajan, K. et al. (2010) Ack1 mediated AKT/PKB tyrosine 176 phosphorylation regulates its activation. *PLoS One* 5, e9646.

Unparalleled Product
Quality, Validation,
and Technical Support

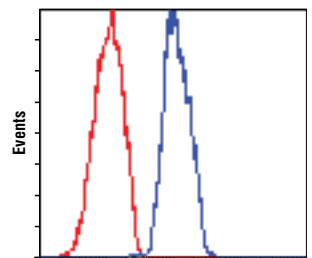
Translational Control

		Applications	Reactivity
#2855	Phospho-4E-BP1 (Thr37/46) (236B4) Rabbit mAb	CA W, IHC-P, IF-IC, F	H, M, R, Mk, Dm
#3929	Phospho-4E-BP1 (Thr37/46) (236B4) Rabbit mAb (Biotinylated)	W, F	H, M, R, Mk, Dm
#4923	Non-phospho-4E-BP1 (Thr46) (87D12) Rabbit mAb	W, F	H, M, R, Mk
#9644	4E-BP1 (53H11) Rabbit mAb	W, IP, IHC-P, IF-IC, F	H, M, R, Mk
#2845	4E-BP2 Antibody	W, IP, IHC-P, F	H, M, R, Mk, B
#2119	BRF1/2 Antibody	W, IF-IC, F	H, M, R, Mk, (C, B)
#9407	DDX6/RCK Antibody	W, IF-IC, F	H, M, R, Mk
#2469	eIF4G (C45A4) Rabbit mAb	W, IHC-P, IF-IC, F	H, M, R, Mk
#2498	eIF4G Antibody	W, IHC-P, IF-IC, F	H, M, R, Mk
XP #5169	eIF4G2/p97 (D88B6) XP® Rabbit mAb	W, IP, IF-IC, F	H, M, R, Mk
XP #5033	ERp72 (D70D12) XP® Rabbit mAb	W, IF-IC, F	H, M, R, Mk
#4675	hnRNP K (R332) Antibody	W, IP, IF-IC, F	H, M, R, Mk
#2983	mTOR (7C10) Rabbit mAb	CA W, IHC-P, IF-IC, F	H, M, R, Mk
#9480	RagC (D8H5) Rabbit mAb	W, IP, IF-IC, F	H, M, R, Mk
New #6960	RCAS1 Antibody	W, IP, IF-IC, F	H, M, R, Mk
#4935	Rheb Antibody	W, IHC-P, F	H, M, R, Sc, (B)
XP #4858	Phospho-S6 Ribosomal Protein (Ser235/236) (D57.2.2E) XP® Rabbit mAb	CA W, IHC-P, IHC-F, IF-IC, F	H, M, R, Mk, Sc, (C)
XP #3945	Phospho-S6 Ribosomal Protein (Ser235/236) (D57.2.2E) XP® Rabbit mAb (Biotinylated)	W, F	H, M, R, Mk, Sc
#4856	Phospho-S6 Ribosomal Protein (Ser235/236) (2F9) Rabbit mAb	CA W, IF-IC, F	H, M, R, Mk
#2211	Phospho-S6 Ribosomal Protein (Ser235/236) Antibody	W, IP, IHC-P, IHC-F, IF-IC, F	H, M, R, Mk, Sc, (C, X)
XP #5364	Phospho-S6 Ribosomal Protein (Ser240/244) (D68F8) XP® Rabbit mAb	CA W, IP, IHC-P, IF-IC, F	H, M, R, Mk
#4278	Phospho-S6 Ribosomal Protein (Ser240/244) (61H9) Rabbit mAb (Biotinylated)	W, F	H, M, R, Mk
#2317	S6 Ribosomal Protein (54D2) Mouse mAb	CA W, IP, IHC-P, IF-IC, F	H, M, R, Mk, Dm
#3944	S6 Ribosomal Protein (54D2) Mouse mAb (Biotinylated)	W, F	H, M, R, Mk
New XP #8509	TIAR (D32D3) XP® Rabbit mAb	W, IP, IF-IC, F	H, M, R, Mk



Phospho-4E-BP1 (Thr37/46) (PE Conjugate)

Phospho-4E-BP1 (Thr37/46) (236B4) Rabbit mAb (PE Conjugate) #7547: Flow cytometric analysis of Jurkat cells, untreated (green) or treated with LY294002 #9901, Wortmannin #9951, and U0126 #9903 (blue), using #7547.



mTOR (Alexa Fluor® 647 Conjugate)

mTOR (7C10) Rabbit mAb (Alexa Fluor® 647 Conjugate) #5048: Flow cytometric analysis of HeLa cells using #5048 (blue) compared to concentration-matched Rabbit (DA1E) mAb IgG XP® Isotype Control (Alexa Fluor® 647 Conjugate) #2985 (red).

Product References:

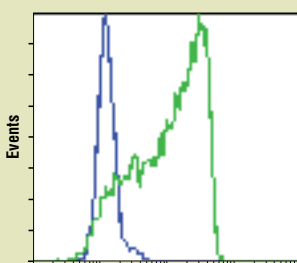
Phospho-S6 Ribosomal Protein (Ser235/236) (2F9) Rabbit mAb #4856: Kotecha, N. et al. (2008) Single-cell profiling identifies aberrant STAT5 activation in myeloid malignancies with specific clinical and biologic correlates. *Cancer Cell* 14, 335–343.

Phospho-S6 Ribosomal Protein (Ser235/236) (D57.2.2E) XP® Rabbit mAb (Alexa Fluor® 647 Conjugate) #4851: Soond, D.R. et al. (2010) PI3K p110delta regulates T-cell cytokine production during primary and secondary immune responses in mice and humans. *Blood* 115, 2203–2213. / Kalland, M.E. et al. (2011) T cell-signaling network analysis reveals distinct differences between CD28 and CD2 costimulation responses in various subsets and in the MAPK pathway between resting and activated regulatory T cells. *J. Immunol.* 187, 5233–5245.

S6 Ribosomal Protein

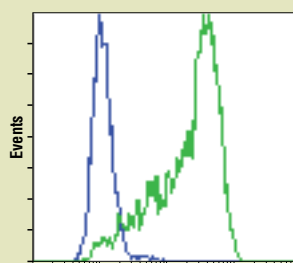
One way that growth factors and mitogens effectively promote sustained cell growth and proliferation is by upregulating mRNA translation. Growth factors and mitogens induce the activation of p70 S6 kinase and the subsequent phosphorylation of the S6 ribosomal protein. Phosphorylation of S6 ribosomal protein correlates with an increase in translation of mRNA transcripts that contain an oligopyrimidine tract in their 5' untranslated regions.

These particular mRNA transcripts (5'TOP) encode proteins involved in cell cycle progression as well as ribosomal proteins and elongation factors necessary for translation. Important S6 ribosomal protein phosphorylation sites include several residues (Ser235, Ser236, Ser240, and Ser244) located within a small, carboxy-terminal region of the S6 protein.



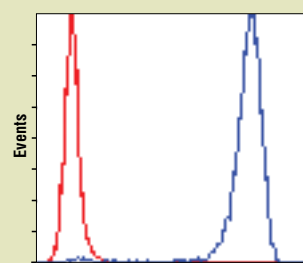
Phospho-S6 Ribosomal Protein (Ser240/244) (Alexa Fluor® 647 Conjugate)

Phospho-S6 Ribosomal Protein (Ser240/244) (D68F8) XP® Rabbit mAb (Alexa Fluor® 647 Conjugate) #5044: Flow cytometric analysis of Jurkat cells, untreated (green) or treated with LY294002 #9901, Wortmannin #9951 and U0126 #9903 (blue), using #5044.



Phospho-S6 Ribosomal Protein (Ser235/236) (Alexa Fluor® 647 Conjugate)

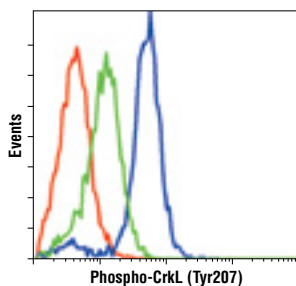
Phospho-S6 Ribosomal Protein (Ser235/236) (D57.2.2E) XP® Rabbit mAb (Alexa Fluor® 647 Conjugate) #4851: Flow cytometric analysis of K-562 cells, untreated (green) or treated with imatinib #9084 (blue), using #4851.



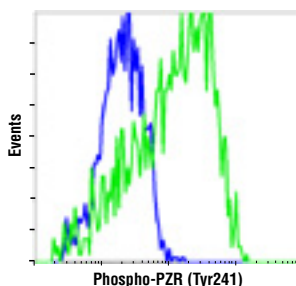
S6 Ribosomal Protein (54D2) Mouse mAb (Alexa Fluor® 488 Conjugate)

S6 Ribosomal Protein (54D2) Mouse mAb (Alexa Fluor® 488 Conjugate) #5317: Flow cytometric analysis of Jurkat cells using #5317 (blue) compared to concentration-matched Mouse (MOPC-21) mAb IgG1 Isotype Control (Alexa Fluor® 488 Conjugate) #4878 (red).

Tyrosine Kinases



Phospho-CrkL (Tyr207) Antibody #3181: Flow cytometric analysis of K-562 cells, untreated (blue) or treated with imatinib #9084 (green), using #3181 compared to a concentration-matched nonspecific negative control antibody (red).

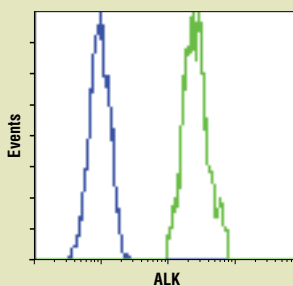


Phospho-PZR (Tyr241) (D6F9) Rabbit mAb #8131: Flow cytometric analysis of BAE cells, untreated (blue) or H₂O₂-treated (green), using #8131.

		Applications	Reactivity
New XP	#3633 ALK (D5F3) XP [®] Rabbit mAb	W, IP, IHC-P, IF-IC, F	H
	#4566 Axl (C44G1) Rabbit mAb	W, IP, F	H, Mk
	#3901 Phospho-Bcr (Tyr177) Antibody	W, IHC-P, F	H, M
	#3902 Bcr Antibody	W, IF-IC, F	H, M, R, Pg
	#3181 Phospho-CrkL (Tyr207) Antibody	W, IP, IHC-P, F	H, M, R, Mk
	#3155 Phospho-M-CSF Receptor (Tyr723) (49C10) Rabbit mAb	W, IP, IHC-P, F	H, M
XP	#3777 Phospho-EGF Receptor (Tyr1068) (D7A5) XP [®] Rabbit mAb	W, IHC-P, IF-IC, F	H, M, R, Mk
XP	#2085 EGF Receptor (E746-A750del Specific) (6B6) XP [®] Rabbit mAb	W, IP, IHC-P, IF-IC, IF-P, F	H
	#3197 EGF Receptor (L858R Mutant Specific) (43B2) Rabbit mAb	W, IP, IHC-P, IF-IC, IF-P, F	H
XP	#4267 EGF Receptor (D38B1) XP [®] Rabbit mAb	CA W, IP, IHC-P, IF-IC, F	H, M, Mk
	#2736 Fes Antibody	W, IP, F	H
	#2755 Fgr Antibody	W, IP, F	H
	#2165 HER2/ErbB2 (29D8) Rabbit mAb	W, IP, IHC-P, IHC-F, IF-IC, F	H, (M, R)
	#3308 c-Kit (Ab81) Mouse mAb	CA W, IP, IF-IC, F	H
New XP	#8198 Met (D1C2) XP [®] Rabbit mAb	CA W, IP, IHC-P, IHC-F, IF-IC, F	H
New	#5631 Met (11C4) Mouse mAb (Flow Specific)	F	H
XP	#5241 PDGF Receptor α (D13C6) XP [®] Rabbit mAb	W, IHC-P, IF-IC, F	H
XP	#3174 PDGF Receptor α (D1E1E) XP [®] Rabbit mAb	W, IP, IHC-P, IF-IC, F	H, M
	#3164 PDGF Receptor α Antibody	W, IP, IHC-P, F	H, M, R
	#3161 Phospho-PDGF Receptor β (Tyr751) Antibody	W, F	H, M, R
	#4564 PDGF Receptor β (C82A3) Rabbit mAb	W, IHC-P, F	H, M, R
New	#8131 Phospho-PZR (Tyr241) (D6F9) Rabbit mAb	W, IP, IF-IC, F	H, M, R, B
New	#8088 Phospho-PZR (Tyr263) (D6A5) Rabbit mAb	W, IP, IF-IC, F	H, M, R, B
	#3223 Ret (C31B4) Rabbit mAb	W, IP, IF-IC, F	H
	#2093 SCF (C19H6) Rabbit mAb	W, IHC-P, F	H
	#2109 Src (36D10) Rabbit mAb	W, IP, IHC-P, IHC-F, IF-F, IF-IC, F	H, M, R, Hm, Mk, B, Pg, (C)
	#2108 Src Antibody	W, IP, IF-IC, F	H, M, R, Mk, (C)

ALK

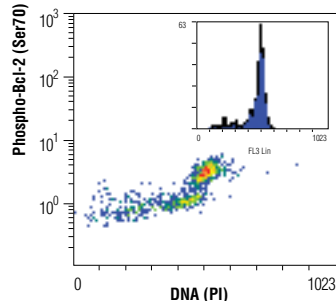
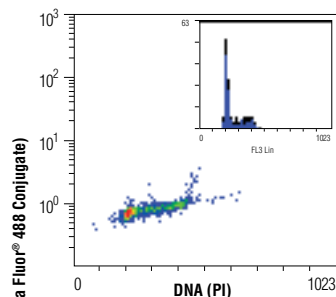
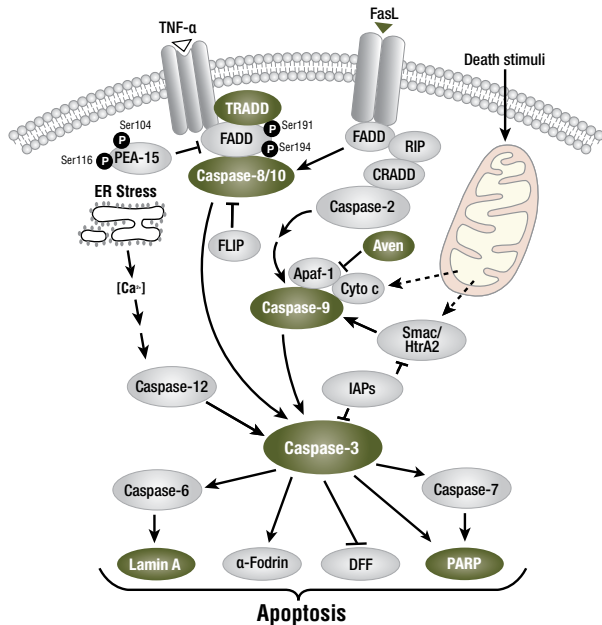
Anaplastic lymphoma kinase (ALK) is a tyrosine kinase receptor for pleiotrophin (PTN), a growth factor involved in embryonic brain development¹⁻³. In ALK-expressing cells, PTN induces phosphorylation of both ALK and the downstream effectors IRS-1, Shc, PLCγ, and PI3 kinase¹. ALK was originally discovered as a nucleophosmin (NPM)-ALK fusion protein produced by a translocation⁴. Investigators have found that the NPM-ALK fusion protein is a constitutively active, oncogenic tyrosine kinase associated with anaplastic lymphoma⁴. Research literature suggests that activation of PLCγ by NPM-ALK may be a crucial step for its mitogenic activity and involved in the pathogenesis of anaplastic lymphomas⁵. A distinct ALK oncogenic fusion protein involving ALK and echinoderm microtubule-associated protein like 4 (EML4) has been described in the research literature from a non-small cell lung cancer (NSCLC) cell line, with corresponding fusion transcripts present in some cases of lung adenocarcinoma. The EML4-ALK fusion protein has been found in 3-7% of NSCLC samples⁶⁻¹⁴.



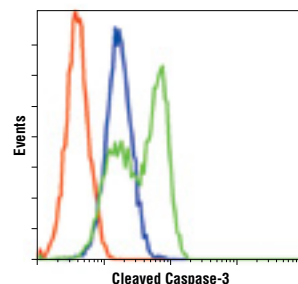
ALK (D5F3) XP[®] Rabbit mAb #3633: Flow cytometric analysis of DU 145 (blue) and KARPAS-299 (green) cells using #3633.

- 1 Stoica, G.E. et al. (2001) *J. Biol. Chem.* 276, 16772-16779.
- 2 Iwahara, T. et al. (1997) *Oncogene* 14, 439-449.
- 3 Morris, S.W. et al. (1997) *Oncogene* 14, 2175-2188.
- 4 Morris, S.W. et al. (1994) *Science* 263, 1281-1284.
- 5 Bai, R.Y. et al. (1998) *Mol. Cell. Biol.* 18, 6951-6961.
- 6 Rikova, K. et al. (2007) *Cell* 131, 1190-1203.
- 7 Takeuchi, K. et al. (2008) *Clin. Cancer Res.* 14, 6618-6624.
- 8 Soda, M. et al. (2007) *Nature* 448, 561-566.
- 9 Takeuchi, K. et al. (2009) *Clin. Cancer Res.* 15, 3143-3149.
- 10 Palmer, R.H. et al. (2009) *Biochem. J.* 420, 345-361.
- 11 Horn, L. and Pao, W. (2009) *J. Clin. Oncol.* 27, 4232-4235.
- 12 Rodig, S.J. et al. (2009) *Clin. Cancer Res.* 15, 5216-5223.
- 13 Mino-Kenudson, M. et al. (2010) *Clin. Cancer Res.* 16, 1561-1571.
- 14 Kwak, E.L. et al. (2010) *N. Engl. J. Med.* 363, 1693-1703.

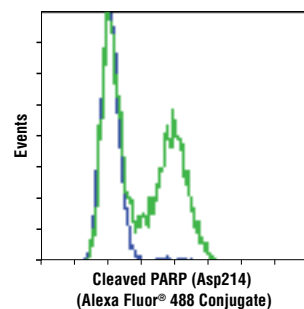
Apoptosis and Autophagy



Phospho-Bcl-2 (Ser70) (5H2) Rabbit mAb (Alexa Fluor® 488 Conjugate) #2834: Flow cytometric analysis of Jurkat cells, untreated (upper) or treated with paclitaxel #9807 (lower), using #2834 and Propidium iodide (PI) #4087. The blue inserts represent propidium iodide staining alone, showing cells in mitotic arrest in the paclitaxel-treated sample.



Cleaved Caspase-3 (Asp175) (5A1E) Rabbit mAb #9664: Flow cytometric analysis of Jurkat cells, untreated (blue) or etoposide-treated (green), using #9664 compared to a concentration-matched nonspecific negative control antibody (red).



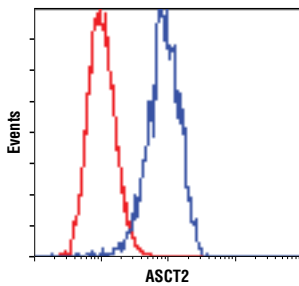
Cleaved PARP (Asp214) (D64E10) XP® Rabbit mAb (Alexa Fluor® 488 Conjugate) #9148: Flow cytometric analysis of Jurkat cells, untreated (blue) or etoposide-treated (green), using #9148.

		Applications	Reactivity
	#2300 Aven Antibody	W, IF-IC, F	H, M, R, Mk
	#5284 Phospho-Bad (Ser112) (40A9) Rabbit mAb	W, IHC-P, F	H, M, R, Mk
	#9291 Phospho-Bad (Ser112) Antibody	W, IP, F, E-P	H, M, R, Mk
	#2827 Phospho-Bcl-2 (Ser70) (5H2) Rabbit mAb	CA W, IF-IC, F	H
	#2764 Bcl-xL (54H6) Rabbit mAb	CA W, IP, IHC-P, IHC-F, IF-IC, F	H, M, R, Mk
	#2933 Bim (C34C5) Rabbit mAb	W, IP, IHC-P, IF-IC, F	H, M, R, (Mk, B, Dg)
	#2819 Bim Antibody	W, IP, IF-IC, F	H, M, R, (Mk)
New	#9579 Cleaved Caspase-3 (Asp175) (D3E9) Rabbit mAb	IHC-P, IF-IC, F	H, (M, R, Mk, B, Pg)
	#9664 Cleaved Caspase-3 (Asp175) (5A1E) Rabbit mAb	CA W, IP, IHC-P, IHC-F, IF-IC, F	H, M, R, Mk, (Dg)
	#9661 Cleaved Caspase-3 (Asp175) Antibody	CA W, IP, IHC-P, IHC-F, IF-IC, F	H, M, R, Mk, (B, Dg, Pg)
New	#8438 Cleaved Caspase-7 (Asp198) (D6H1) Rabbit mAb	W, IF-IC, F	H, M, R, (Mk)
New XP®	#8592 Cleaved Caspase-8 (Asp387) (D5B2) XP® Rabbit mAb (Mouse Specific)	W, IP, IF-IC, F	M
	#9496 Cleaved Caspase-8 (Asp391) (18C8) Rabbit mAb	W, IHC-P, IF-IC, F	H
	#9502 Caspase-9 Antibody (Human Specific)	W, F	H
New	#8023 Fas (4C3) Mouse mAb	W, IP, IF-IC, F	H
	#4777 Lamin A/C (4C11) Mouse mAb	CA W, IP, IHC-P, IF-F, IF-IC, F	H, M, R, Mk
XP®	#4599 LC3A (D50G8) XP® Rabbit mAb	W, IP, IHC-P, IF-IC, F	H, M, R, (Mk, Dg)
	#4108 LC3A/B Antibody	W, IF-IC, F	H, M, R, (Mk, C, X, Z, Dg)
XP®	#3868 LC3B (D11) XP® Rabbit mAb	W, IP, IHC-P, IF-IC, F	H, M, R, (Mk, B, Pg)
	#2775 LC3B Antibody	W, IF-IC, F	H, M, R, (Mk, B, Pg)
New	#9891 NBR1 (D2E6) Rabbit mAb	W, IP, IF-IC, F	H, M
XP®	#5605 c-Myc (D84C12) XP® Rabbit mAb	W, IP, IF-IC, F	H, M, R, (Mk, Dg, Pg)
	#2328 PAR-4 Antibody	W, IP, IF-IC, F	H, M, R, Mk
XP®	#5625 Cleaved PARP (Asp214) (D64E10) XP® Rabbit mAb	CA W, IP, IHC-P, IF-IC, F	H, Mk
	#9541 Cleaved PARP (Asp214) Antibody (Human Specific)	W, IHC-P, IF-IC, F	H
	#3693 Perforin Antibody (Mouse Specific)	W, IF-IC, F	M
	#2808 Survivin (71G4B7) Rabbit mAb	CA W, IP, IHC-P, IHC-F, IF-IC, F	H, M, R
	#4037 Survivin (71G4B7) Rabbit mAb (Biotinylated)	W, F	H, M, R
	#3694 TRADD Antibody	W, IP, F	H, M, R, Mk
	#4715 TRAF1 (45D3) Rabbit mAb	W, IP, IHC-P, IF-IC, F	H, (Mk)
	#3219 TRAIL (C92B9) Rabbit mAb	W, IP, IHC-P, IF-IC, F	H

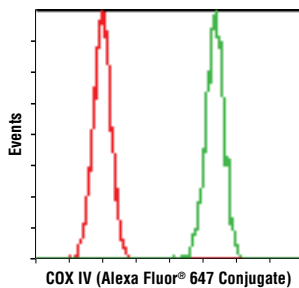
Product References:

Bim Antibody #2819: Kurtulus, S. et al. (2011) Bcl-2 allows effector and memory CD8+ T cells to tolerate higher expression of Bim. *J. Immunol.* 186, 5729-5737.

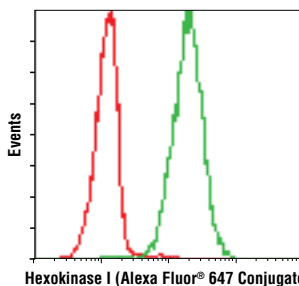
Metabolism



ASCT2 (D7C12) Rabbit mAb #8057: Flow cytometric analysis of HT-29 cells using #8057 (blue) compared to concentration-matched Rabbit (DA1E) mAb IgG XP® Isotype Control #3900 (red).



COX IV (3E11) Rabbit mAb (Alexa Fluor® 647 Conjugate) #7561: Flow cytometric analysis of HeLa cells using #7561 (green) compared to concentration-matched Rabbit (DA1E) mAb IgG XP® Isotype Control (Alexa Fluor® 647 Conjugate) #2985 (red).



Hexokinase I (C35C4) Rabbit mAb (Alexa Fluor® 647 Conjugate) #3540: Flow cytometric analysis of MCF7 cells using #3540 (green) compared to concentration-matched Rabbit (DA1E) mAb IgG XP® Isotype Control (Alexa Fluor® 647 Conjugate) #2985 (red).

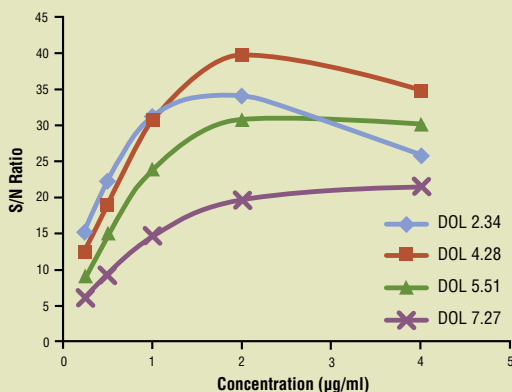
Unparalleled Product
Quality, Validation,
and Technical Support

	Applications	Reactivity
#3676 Acetyl-CoA Carboxylase (C83B10) Rabbit mAb	W, IHC-P, IF-IC, F	H, M, R, Hm
#3662 Acetyl-CoA Carboxylase Antibody	W, IP, IHC-P, IF-IC, F	H, M, R, Mk, B, (C, Dm)
#4150 AMPKβ1/2 (57C12) Rabbit mAb	W, IHC-P, IF-IC, F	H, M, R, Hm, Mk
New #8057 ASCT2 (D7C12) Rabbit mAb	W, IP, IF-IC, F	H, M, Mk
#5345 ASCT2 (V501) Antibody	W, IP, IF-IC, F	H, M, R
New XP #5864 CA12 (D75C6) XP® Rabbit mAb	W, IP, F	H
#4850 COX IV (3E11) Rabbit mAb	CA W, IP, IHC-P, IHC-F, IF-IC, F	H, R, Mk, Z, B, Pg
#4844 COX IV Antibody	W, IP, IHC-P, IF-IC, F	H, M, R, Mk, B
#2118 GAPDH (14C10) Rabbit mAb	CA W, IHC-P, IF-IC, F	H, M, R, Mk
#5014 GAPDH (14C10) Rabbit mAb (Biotinylated)	W, IF-IC, F	H, M, R, Mk
#3893 Glycogen Synthase Antibody	W, IP, F	H, M, R
#3689 Hexokinase I (C35C4) Rabbit mAb (Alexa Fluor® 488 Conjugate)	IF-IC, F	H, M
#3540 Hexokinase I (C35C4) Rabbit mAb (Alexa Fluor® 647 Conjugate)	IF-IC, F	H, M
New #8137 IDH1 (D2H1) Rabbit mAb	W, IP, IF-IC, F	H, M, R, Mk
New XP #9750 IGF-1 Receptor β (D23H3) XP® Rabbit mAb	W, IP, IF-IC, F	H, M, R, Mk
#3014 Insulin (C27C9) Rabbit mAb	IHC-P, IF-F, IF-IC, F	H, M, R
#4590 Insulin Antibody	IHC-P, IF-F, IF-IC, F	H, M, R

Custom Conjugation

Cell Signaling Technology (CST) offers custom conjugation of our highest quality antibodies to Alexa Fluor® dyes, R-Phycoerythrin (PE), sepharose or magnetic beads, as well as biotin or HRP. This service includes research application consultation and extensive technical support from our Custom Conjugation Group. Contact your local sales representative for more information and multiple sizing options.

Custom conjugations are optimized by degree of labeling (DOL) testing to identify the optimal antibody: dye molecule ratio, resulting in conjugates with maximum signal intensity.

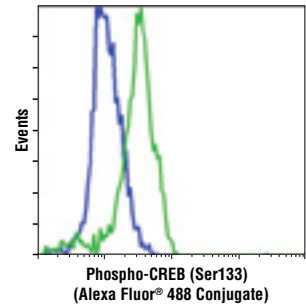


- All CST™ custom conjugates are validated in the intended application.
- CST™ antibodies can be conjugated to Alexa Fluor® 488, 555, 594, or 647, PE, sepharose or magnetic beads, biotin, or HRP.
- CST™ monoclonal antibodies, including XP® monoclonal antibodies, yield highly reproducible results, minimizing lot-to-lot variation.

Other Antibodies

Calcium, cAMP, and Lipid Signaling

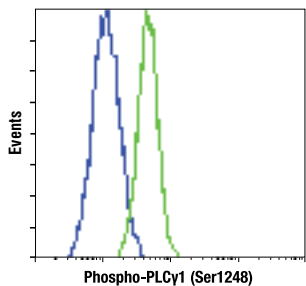
		Applications	Reactivity
New	#8235 Annexin A2 (D11G2) Rabbit mAb	W, IHC-P, IF-IC, F	H, M, R, Mk, B, Pg, (Dg)
	#9198 Phospho-CREB (Ser133) (87G3) Rabbit mAb	CA W, IHC-P, IHC-F, IF-F, IF-IC, F, ChIP	H, M, R
XP	#5607 MARCKS (D88D11) XP [®] Rabbit mAb	W, IHC-P, IF-IC, F	H, Mk
	#9694 PIP5K2B Antibody	W, F	H, M, R, Mk
	#4782 PKA C- α Antibody	W, IP, IF-IC, F	H, M, R
	#2056 PKC α Antibody	W, IP, IF-IC, F	H, M, R, Mk, (Dg)
	#2998 PKC ζ (C83H11) Rabbit mAb	W, F	H, M, R, Mk
	#2052 PKD/PKC μ Antibody	W, F	H, M, R, Mk
	#2821 Phospho-PLC γ 1 (Tyr783) Antibody	W, F	H, M, R
New	#8713 Phospho-PLC γ 1 (Ser1248) (D25A9) Rabbit mAb	W, IP, IHC-P, IF-IC, F	H, M, Mk, (R)
New	#5529 S100A10 (4E7E10) Mouse mAb	W, IHC-P, IF-IC, F	H, Mk



Phospho-CREB (Ser133) (87G3) Rabbit mAb (Alexa Fluor[®] 488 Conjugate) #9187: Flow cytometric analysis of NIH/3T3 cells, untreated (blue) or treated with hPDGF-BB #8912 (green), using #9187.

Nuclear Receptor Signaling

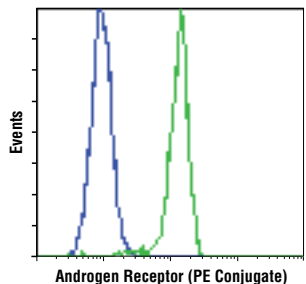
XP	#5153 Androgen Receptor (D6F11) XP [®] Rabbit mAb	CA W, IHC-P, IF-IC, F	H
XP	#3960 Nur77 (D63C5) XP [®] Rabbit mAb	W, IP, IF-IC, F	H, (Mk)
	#3157 Progesterone Receptor B (C1A2) Rabbit mAb	W, IHC-P, IF-IC, F	H
	#2115 SRC-3 (11B1) Mouse mAb	W, IF-IC, F	H



Phospho-PLC γ 1 (Ser1248) (D25A9) Rabbit mAb #8713: Flow cytometric analysis of Jurkat cells, treated with U0126 #9903 (blue) or TPA #4174 (green), using #8713.

Phosphatases

	#2614 Pan-Calcieneurin A Antibody	W, IP, IF-IC, F	H, M, R, Mk, Dm, (C, X, B, Pg)
	#2039 PP2A A Subunit Antibody	W, IF-IC, F	H, M, R, Mk, Dm
	#4953 PP2A B Subunit Antibody	W, IP, IHC-P, IF-IC, F	H, M, R, Mk, (C)
	#2259 PP2A C Subunit (52F8) Rabbit mAb	W, IP, IHC-P, F	H, M, R, Mk, Dm, (C, Pg)
	#2038 PP2A C Subunit Antibody	W, IP, IHC-P, IF-IC, F	H, M, R, Mk, Dm, (C, Pg)
XP	#3549 PP2C- α (D18C10) XP [®] Rabbit mAb	W, IP, IHC-P, IF-IC, F	H, Mk
	#2289 PP5 Antibody	W, F	H, M, R, Mk
	#2725 SHIP1 (C15C9) Rabbit mAb	W, IP, F	H
	#2727 SHIP1 (C40G9) Rabbit mAb	W, IP, IF-IC, F	H
	#2726 SHIP1 (P290) Antibody	W, IP, IF-IC, F	H
XP	#5445 Phospho-SHIP2 (Tyr1135) (D33C6) XP [®] Rabbit mAb	W, IP, F	H
	#2839 SHIP2 (C76A7) Rabbit mAb	W, IP, IF-IC, F	H
	#5431 Phospho-SHP-2 (Tyr580) (D66F10) Rabbit mAb	W, IP, F	M, R, (H)



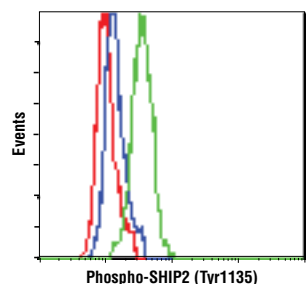
Androgen Receptor (D6F11) XP[®] Rabbit mAb (PE Conjugate) #8428: Flow cytometric analysis of Jurkat (blue) and LNCaP (green) cells using #8428.

Phospho-Ser/Thr/Tyr Motif Antibodies

#9411 Phospho-Tyrosine Mouse mAb (P-Tyr-100)	CA W, IP, IHC-P, IF-F, IF-IC, All IF-P, F, E-P
#9416 Phospho-Tyrosine Mouse mAb (P-Tyr-102)	W, IP, IHC-P, F, E-P All

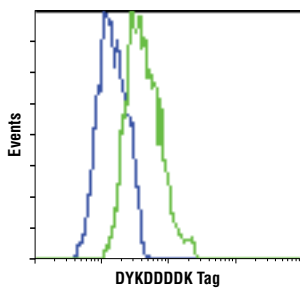
Product References:

Phospho-Tyrosine Mouse mAb (P-Tyr-100) (Alexa Fluor[®] 647 Conjugate) #9415: Kalland, M.E. et al. (2011) T cell signaling network analysis reveals distinct differences between CD28 and CD2 costimulation responses in various subsets and in the MAPK pathway between resting and activated regulatory T cells. *J. Immunol.* 187, 5233–5245.

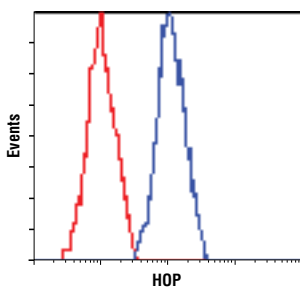


Phospho-SHIP2 (Tyr1135) (D33C6) XP[®] Rabbit mAb #5445: Flow cytometric analysis of K-562 cells, untreated (green) or λ phosphatase-treated (blue), using #5445 compared to concentration-matched Rabbit (DA1E) mAb IgG XP[®] Isotype Control #3900 (red).

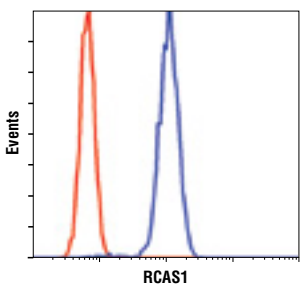
Other Antibodies Continued



DYKDDDDK Tag (9A3) Mouse mAb (Binds to same epitope as Sigma's Anti-FLAG® M2 Antibody) #8146: Flow cytometric analysis of COS-7 cells, untreated (blue) or transfected with a construct expressing DYKDDDDK-tagged IRF3 (green), using #8146.



HOP (D10E2) XP® Rabbit mAb #5670: Flow cytometric analysis of HeLa cells using #5670 (blue) compared to concentration-matched Rabbit (DA1E) mAb IgG XP® Isotype Control #3900 (red).



RCAS1 Antibody #6960: Flow cytometric analysis of Jurkat cells using #6960 (blue) compared to concentration-matched Rabbit (DA1E) mAb IgG XP® Isotype Control #3900 (red).

Epitope Tag Antibodies

		Applications	Reactivity
New	#8146 DYKDDDDK Tag (9A3) Mouse mAb (Binds to same epitope as Sigma's Anti-FLAG® M2 Antibody)	W, IP, IHC-P, IF-IC, F	All
	#2372 β-Gal (14B7) Mouse mAb	W, IP, IF-IC, F	All
XP®	#2956 GFP (D5.1) XP® Rabbit mAb	W, IHC-P, IF-IC, F	All
	#2955 GFP (4B10) Mouse mAb	W, IHC-P, F	All
	#3724 HA-Tag (C29F4) Rabbit mAb	W, IP, IHC-P, IF-IC, F	All
	#5017 HA-Tag (C29F4) Rabbit mAb (Biotinylated)	W, F	All
	#2367 HA-Tag (6E2) Mouse mAb	W, IHC-P, IF-IC, F	All
	#3368 GST (26H1) Mouse mAb (Alexa Fluor® 488 Conjugate)	IF-IC, F	All
	#3445 GST (26H1) Mouse mAb (Alexa Fluor® 647 Conjugate)	F	All
	#2278 Myc-Tag (71D10) Rabbit mAb	W, IP, IF-IC, F	All
	#3946 Myc-Tag (71D10) Rabbit mAb (Biotinylated)	W, F	All
	#2276 Myc-Tag (9B11) Mouse mAb	W, IP, IHC-P, IF-IC, F	All
	#2272 Myc-Tag Antibody	W, IF-IC, F	All

Protein Folding and Stability

	#2539 Calpain 2 Large Subunit (M-type) Antibody	W, IP, IF-F, F	H, M, R
	#2284 Cathepsin D Antibody	W, IHC-P, F	H, Mk
New XP®	#5670 HOP (D10E2) XP® Rabbit mAb	W, IF-IC, F	H, Mk, B
	#4356 HSF1 Antibody	W, IP, IHC-P, IF-IC, F, ChIP	H, M, R, Mk
	#2405 Phospho-HSP27 (Ser78) Antibody	W, IHC-P, F	H, Mk
	#2406 Phospho-HSP27 (Ser82) Antibody II	W, IHC-P, IHC-F, IF-IC, F	H, M, R, Mk
	#2442 HSP27 Antibody (Rodent Preferred)	W, IF-IC, F	M, R
	#4868 HSP40 Antibody	W, IP, IF-IC, F	H, M, R, Mk
	#4870 HSP60 (D307) Antibody	W, IF-IC, F	H, M, R, Mk, Dm
	#4869 HSP60 (D85) Antibody	W, IF-IC, F	H, M, R, Mk, Dm
	#4876 HSP70 (D69) Antibody	W, IHC-P, IHC-F, IF-IC, F	H, M, R, Mk
	#4873 HSP70 (6B3) Rat mAb	W, IP, IHC-P, IF-IC, F	H, Mk
	#4877 HSP90 (C45G5) Rabbit mAb	W, IHC-P, IF-IC, F	H, M, R, Mk, (B)
	#4875 HSP90 (E289) Antibody	W, IHC-P, F	H, M, R, Mk, (B)
	#2743 ISG15 Antibody	W, F, E-P	H, M, Mk
	#2740 NEDD4 Antibody	W, IF-IC, F	H, M, R, Mk
	#2754 NEDD8 (19E3) Rabbit mAb	W, IP, IHC-P, F	H, M, R, Mk, (X, Z, B)
	#2745 NEDD8 Antibody	W, IP, IHC-P, F	H, M, R, Mk, (X, Z, B)
New	#6960 RCAS1 Antibody	W, IP, IF-IC, F	H, M, R, Mk
	#2156 Skp1 Antibody	W, IF-IC, F	H, M, R
	#4997 UBC3 Antibody	W, F	H, M, R
	#4996 UBC3B Antibody	W, IHC-P, F	H, M, R
	#4890 UBE1a Antibody	W, IP, IHC-P, IF-IC, F	H, M, R
	#2648 VCP Antibody	W, IF-IC, F	H, M, R, Mk, (X, Z, B, Pg, Sc)

Product References:

HA-Tag (6E2) Mouse mAb (Alexa Fluor® 488 Conjugate) #2350: Mahajan, K. et al. (2010) Ack1 mediated AKT/PKB tyrosine 176 phosphorylation regulates its activation. *PLoS One* 5, e9646.

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Flow Cytometry Protocols

Cell Signaling Technology recommends using the associated optimized protocol for each of our flow cytometry validated antibodies. Please find a link to the appropriate protocol from the product webpage on our website.

General Flow Cytometry Protocol

A. Solutions and Reagents

NOTE: Prepare solutions with Milli-Q® or equivalently purified water.

- 1. 1X Phosphate Buffered Saline (PBS):** Dissolve 8 g NaCl, 0.2 g KCl, 1.44 g Na₂HPO₄ and 0.24 g KH₂PO₄ in 800 mL dH₂O. Adjust the pH to 7.4 with HCl and the volume to 1L. Store at room temperature.
- 2. Formaldehyde** (methanol free).
- 3. Incubation Buffer:** Dissolve 0.5 g bovine serum albumin (BSA) in 100mL 1X PBS. Store at 4°C.

B. Fixation

1. Collect cells by centrifugation and aspirate supernatant.
2. Resuspend cells briefly in 0.5-1 ml PBS. Add formaldehyde to a final concentration of 2-4% formaldehyde.
3. Fix for 10 minutes at 37°C.
4. Chill tubes on ice for 1 minute.
5. For extracellular staining with antibodies that do not require permeabilization, proceed to step D1 or store cells in PBS with 0.1% sodium azide at 4°C; for intracellular staining, proceed to permeabilization step C1.

C. Permeabilization

1. Permeabilize cells by adding ice-cold 100% methanol slowly to pre-chilled cells, while gently vortexing, to a final concentration of 90% methanol. Alternatively, to remove fix prior to permeabilization, pellet cells by centrifugation and resuspend in 90% methanol.
2. Incubate 30 minutes on ice.
3. Proceed with staining or store cells at -20°C in 90% methanol.

Protocol for Combined Staining of Intracellular Proteins and Cell Surface Markers in Blood

A. Solutions and Reagents

NOTE: Prepare solutions with Milli-Q® or equivalently purified water.

- 1. 10X Phosphate Buffered Saline (PBS):** To prepare 1 L add 80 g sodium chloride (NaCl), 2 g potassium chloride (KCl), 14.4 g sodium phosphate, dibasic (Na₂HPO₄) and 2.4 g potassium phosphate, monobasic (KH₂PO₄) to 1 L dH₂O. Adjust pH to 7.4. Store at room temperature.
- 2. Formaldehyde** (methanol free).
- 3. Incubation Buffer:** Dissolve 0.5 g bovine serum albumin (BSA) in 100 mL 1X PBS. Store at 4°C.

B. Preparation of Whole Blood (fixation, lysis, and permeabilization) for Immunostaining

1. Aliquot 100 µl fresh whole blood per assay tube.
2. **OPTIONAL:** Place tubes in rack in 37°C water bath for short-term treatments with ligands, inhibitors, drugs, etc.
3. Add 65 µl of 10% formaldehyde to each tube.
4. Vortex briefly and let stand for 15 minutes at room temperature.
5. Add 1 ml of Triton X-100 in PBS to make 0.1% Triton final concentration.
6. Vortex and let stand for 30 minutes at room temperature.
7. Add 1 ml Incubation Buffer.
8. Pellet cells by centrifugation and aspirate supernatant.

D. Immunostaining

NOTE: Account for isotype matched controls for monoclonal antibodies or species matched IgG for polyclonal antibodies. Count cells using a hemocytometer or alternative method.

1. Aliquot 0.5-1x10⁶ cells into each assay tube (by volume).
2. Add 2-3 ml Incubation Buffer to each tube and rinse by centrifugation. Repeat.
3. Resuspend cells in 100 µl Incubation Buffer per assay tube.
4. Block in Incubation Buffer for 10 minutes at room temperature.
5. Add the unconjugated, biotinylated, or fluorochrome-conjugated primary antibody at the appropriate dilution to the assay tubes (see individual antibody datasheet for the appropriate dilution).
6. Incubate for 1 hour at room temperature.
7. Rinse as before in Incubation Buffer by centrifugation.
8. If using a fluorochrome-conjugated primary antibody, resuspend cells in 0.5 ml PBS and analyze on flow cytometer; for unconjugated or biotinylated primary antibodies, proceed to step D9.
9. Resuspend cells in fluorochrome-conjugated secondary antibody* or fluorochrome-conjugated avidin, diluted in Incubation Buffer at the recommended dilution.
10. Incubate for 30 minutes at room temperature.
11. Rinse as before in Incubation Buffer by centrifugation.
12. Resuspend cells in 0.5 ml PBS and analyze on flow cytometer; alternatively, for DNA staining, proceed to step E1.

* Recommended secondary antibodies are listed on page 5.

E. Optional DNA Stain

1. Resuspend cells in 0.5 ml of DNA dye (e.g. DRAQ5® #4084).
2. Incubate for at least 5 minutes at room temperature.
3. Analyze cells in DNA stain on flow cytometer.

9. Resuspend cells in cold 50% methanol in PBS (store methanol solution at -20°C until just before use).

10. Incubate at least 10 minutes on ice.

11. Proceed with staining or store cells at -20°C in 50% methanol.

C. Staining Using Unlabeled Primary and Conjugated Secondary Antibodies

1. Add 1 ml Incubation Buffer to each tube and rinse by centrifugation. Repeat.
2. Add primary antibodies diluted as recommended on datasheet in Incubation Buffer.
3. Incubate for 30-60 minutes at room temperature.
4. Rinse as before in incubation buffer by centrifugation.
5. Resuspend cells in fluorochrome-conjugated secondary antibody* diluted in Incubation Buffer according to the manufacturer's recommendations.
6. Incubate for 30 minutes at room temperature.
7. Rinse as before in Incubation Buffer by centrifugation.
8. Resuspend cells in 0.5 ml PBS and analyze on flow cytometer.

Reference: Chow S, Hedley D, Grom P, Magari R, Jacobberger JW, Shankey TV (2005) Whole blood fixation and permeabilization protocol with red blood cell lysis for flow cytometry of intracellular phosphorylated epitopes in leukocyte subpopulations. *Cytometry A* 67(1), 4-17.



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