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Research Focus

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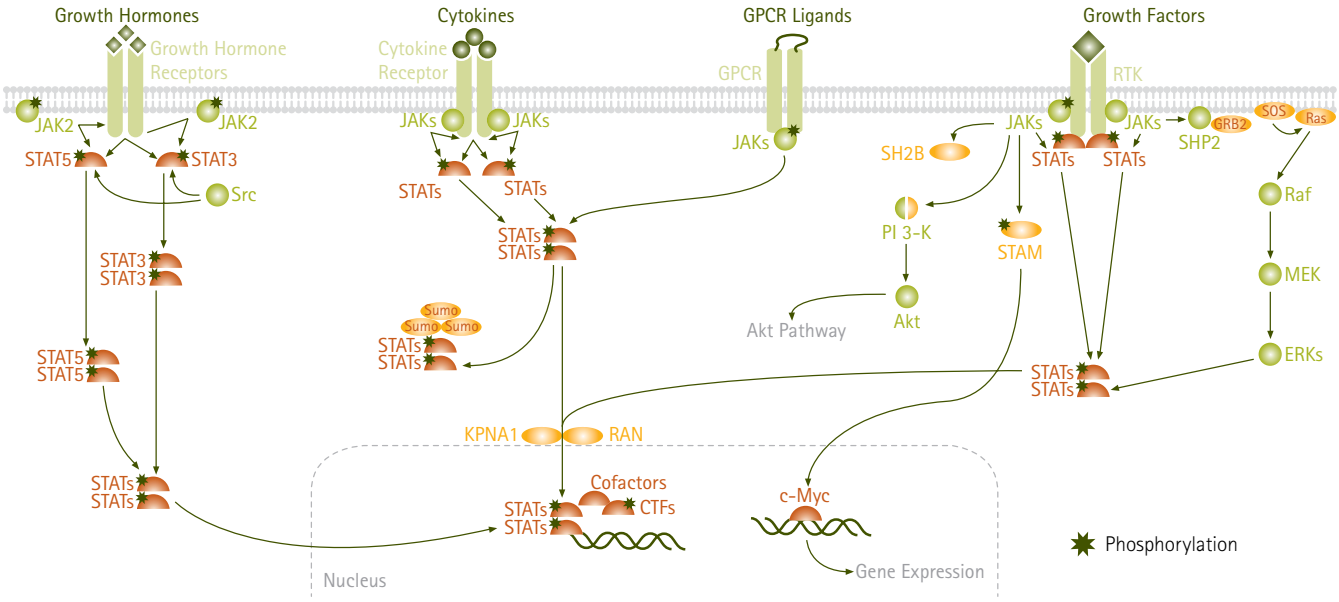
JAK/STAT Signaling

The Janus kinase (JAK)/signal transducers and activators of transcription (STAT) signaling pathway play an important role in cell proliferation, cell differentiation, cell migration, and cell death. It is the principal signaling mechanism for a variety of cytokines and growth factors. Constitutive activation or dysregulation of JAK/STAT signaling can result in inflammatory disease, erythrocytosis, gigantism, and leukemia.

In the JAK/STAT signaling scheme, ligand binding to cytokine receptors induces dimerization of their receptor subunits. They may either form homodimers, as in case of erythropoietin, or heterodimers as in case of interferons and interleukins. Following ligand binding, the cytoplasmic domains of two receptor subunits associate with JAK tyrosine kinases. In mammals, the JAK family is composed of four members: JAK1, JAK2, JAK 3, and Tyk2. Ligand binding brings the two JAKs into close proximity, allowing their transphosphorylation. The activated JAKs can then phosphorylate STATs. In mammalian cells seven different STATs have been recognized. They contain a conserved tyrosine residue near the C-terminus, which is phosphorylated by JAKs. This tyrosine phosphorylation allows the dimerization of STATs by interacting with a



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conserved SH2 domain. In their latent form STATs are located in the cytoplasm and upon phosphorylation they are transported into the nucleus by an importin α-5 dependent mechanism. In the nucleus, dimerized STATs bind to specific regulatory sequences to activate or repress transcription of target genes.

JAK/STAT pathway is reported to play an important role in the establishment of cell fate. For example, IL-12 promotes differentiation of $CD4^+$ T cells to Th1 cells by activating STAT4, and stat4 knockout mice fail to respond to IL-12 and produce only Th2 cells. On the other hand IL-4 activates STAT6 and promotes differentiation of $CD4^+$ T cells to Th2 cells. JAK/STAT pathway is also involved in hematopoietic development. Targeting of Jak2 gene is shown to result in embryonic lethality in mice due to failure of erythropoiesis. In addition, JAK/STAT pathways have been reported to be constitutively active in several forms of human cancers.

References:

Vainchenker, W., and Constantinescu, S.N., 2013. Oncogene 32, 2601.
Yu, H., et al. 2009. Nat. Rev. Cancer 9, 798.
Pfeifer, A.C., et al. 2008. Essays Biochem. 45, 109.
Rawlings, J.S. et al. 2004. J. Cell Sci. 117, 1281.
Shuai, K., and Liu, B. 2003. Nat. Rev. Immunol. 3, 900.

Antibodies for JAK/STAT Signaling Research

Description	Species Reactivity*	Applications**	Qty/Pk	Catalogue No.
Anti-JAK1	H, Ht, M, R, Rb	IP, WB	150 µL	06-272
Anti-JAK 1	H	IP, WB	200 µg	06-665
Anti-JAK1	H, M, R	IP, WB	100 µL	AB3802
Anti-JAK1	H, M, R	WB	100 µg	07-1485
Anti-JAK1, clone 73	H, M, R, Ca, Xn, Ch	IF, IP, WB	100 µg	05-1154
Anti-JAK2	H, M, R	IP, WB	150 µL	06-255
Anti-JAK2	H, M, R	IP, WB	100 µg	06-1310
Anti-JAK2, Agarose conjugate	H, M, R	IP	200 µg	16-121
Anti-JAK2, clone 8E10.2	H, M, R	IH(P), WB	100 µL	04-001
Anti-phospho-JAK2 (Tyr317)	H	WB	100 µL	07-1318
Anti-phospho JAK2 (Tyr570)	H, R, Eq, Chp, Pm, Po	WB	100 µg	09-241
Anti-phospho JAK2 (Tyr813)	H, M, R, Eq, Chp, Po	WB	100 µg	09-242
Anti-phospho-JAK2 (Tyr868)	H, M	WB	100 µL	09-203
Anti-phospho JAK2 (Tyr972)	H, R, Eq, Op, B, Chp, Po	WB	100 µg	09-243
Anti-phospho-JAK2 (Tyr1007/1008)	H, Ch, M, Po, R, Xn, Zebrafish	WB	200 µL	09-275
Anti-phospho-JAK2 (Tyr1007/1008)	H, M, R	WB	200 µg	07-606
Anti-phospho-JAK2 (Tyr1007/Tyr1008), clone E132	H, M, R	ICC, IH(P), IP, WB	100 µL	04-1098
Anti-Phospho-JAK1 (TYR1022/1023)	H, M, R	WB	100 µL	07-1489
Anti-JAK3	M	WB	150 µL	06-342
Anti-JAK3	H, M	IP, WB	100 µL	07-1487
Anti-JAK3	H, M	IH(P), IP, WB	100 µL	07-1488
Anti-JAK3, clone B32-32	H, M	IP	200 µg	05-406
Anti-JAK3, clone HL423	H, M	Mplex, WB	200 µL	04-011
Anti-JAK3, CT	H	WB	250 µg	06-667
Anti-STAT1	H	WB	100 µL	04-069
Anti-STAT1, CT	H, M	EMSA, IP, WB	250 µg	06-501
Anti-STAT1-α	H, M	IP, WB	100 µg	AB16951
Anti-Phospho-STAT1 (Tyr701), clone 5C9.2	H, M, R	WB	100 µg	05-1064

***Species:** Av: avian; B: bovine, Ca: canine; Ch: chicken; Chp: chimpanzee; Eq: equine; H: human; M: mouse, Op: opossum; Pm: primate; R: rat; Sh: sheep; Xn: xenopus
****Applications**: ELISA: enzyme-linked immunosorbent assay; EMSA: electrophoretic mobility shift assay; FC: flow cytometry; ICC: immunocytochemistry, IF: immunofluorescence; IH: immunohistochemistry; IP: immunoprecipitation; Mplex: multiplexing; WB: Western blotting

Antibodies for JAK/STAT Signaling Research (continued)

Description	Species Reactivity*	Applications**	Qty/Pk	Catalogue No.
Anti-phospho-STAT1 (Tyr701)	Ch, H, M, R	WB	100 µL	07-307
Anti-phospho-STAT1 (Tyr701), Alexa Fluor® 488 conjugated, clone 5C9.2	H, M, R	FC, WB	100 tests	FCMAB106A4
Anti-phospho-STAT1 (Ser727)	H, M	WB	200 µL	07-714
Anti-phospho-STAT1 (Ser 727), clone 12C5	H	ELISA, WB	100 µg	04-478
Anti-STAT2	M	ICC, EMSA, IP, WB	200 µg	07-140
Anti-STAT2, clone BK502	H	WB	200 µg	05-693
Anti-STAT2, CT	H, M	ICC, EMSA, IP, WB	200 µg	06-502
Anti-phospho-STAT2 (Tyr689)	H, M	IP, WB	100 µL	ABE541
Anti-phospho-STAT2 (Tyr689)	H, M	IP, WB	200 µL	07-224
Anti-phospho-STAT2 (Tyr972)	H, R, Eq, Op, B, Chp, Po	WB	100 µg	09-243
Anti-STAT3	H, M, R	ICC, EMSA, IP, WB	200 µg	06-596
Anti-STAT3	H, M, R, B, Po, Pm	IP, WB	100 µg	07-2173
Anti-STAT3, clone 18F7.1	H, M	ICC, WB	100 µg	05-1078
Anti-STAT3, clone E121-21	H	ICC, IH(P), IP, WB	100 µL	04-1014
Anti-phospho-STAT3 (Tyr68)	Ch, B, H, M, Po, R, Xn	WB	100 µg	07-1347
Anti-phospho-STAT3 (Tyr705), clone EP2147Y	H, M	ICC, IH(P), IP, WB	100 µL	04-1059
Anti-phospho-STAT3 (Tyr705), clone 9E12	Ca, H, M	ELISA, IP, WB	100 µg	05-485
Anti-phospho-STAT3 (Ser727)	M	WB	100 µL	07-703
Anti-STAT3, phospho (Ser727), clone 6E4	H, M	ICC, IH(P), IP, WB	100 µL	MAB3705
Anti-STAT5, phospho (Tyr694); A & B isoforms	H	IF, WB	50 µg	AB3800
Anti-STAT5A	H, M	WB	200 µg	06-553
Anti-STAT5A	H, M, R	IP, WB	100 µL	06-968
Anti-phospho-STAT5A/B (Ser726/Ser731)	H, M, R	WB	200 µL	06-867
Anti-STAT5A/B	H, M, R, Sh	EMSA	250 µL	06-588
Anti-STAT5B	H, M	WB	200 µg	06-554
Anti-STAT5B	Av, H, M, R	IP, WB	100 µL	06-969
Anti-STAT5α (CT), clone E289	H, R	ICC, FC, IH(P), IP, WB	100 µL	04-1016
Anti-phospho-STAT5A/B (Tyr694/699), clone 8-5-2	B, H, M, R, Sh	WB	100 µg	05-495
Anti-phospho-STAT5A/B (Tyr694/699)	B, H, M, R	DB, WB	100 µL	07-586
Anti-phospho-STAT5A/B (Tyr694/699), clone A11W	B, Chp, H, M, R	WB (Not IP)	100 µL	04-886
Anti-phospho-STAT5A/B (Tyr694/699), clone A11W	H, M	WB	100 µL	05-886
Anti-phospho-STAT5A/B (Ser726/Ser731)	H, M, R	WB	200 µL	06-867
Anti-STAT6	H	WB	100 µg	07-914
Anti-phospho STAT6 (Tyr641), clone 19E2.1	H, M	FC, WB	100 µg	MABE270
Anti-phospho-STAT6 (Tyr641), clone 16E12	H, M, R	WB	100 µg	05-590
Anti-phospho-STAT6 (Tyr641)	H, M	WB	100 µL	06-937

***Species:** Av: avian; B: bovine, Ca: canine; Ch: chicken; Chp: chimpanzee; Eq: equine; H: human; M: mouse, Op: opossum; Pm: primate; R: rat; Sh: sheep; Xn: xenopus

****Applications**: ELISA: enzyme-linked immunosorbent assay; EMSA: electrophoretic mobility shift assay; FC: flow cytometry; ICC: immunocytochemistry, IF: immunofluorescence; IH: immunohistochemistry; IP: immunoprecipitation; Mplex: multiplexing; WB: Western blotting

JAK Inhibitors

Description	Catalogue No.	Comments	Size
JAK Inhibitor I (Pyridone 6)	420099	A cell-permeable, reversible, ATP-competitive inhibitor of JAK1 (IC_{50} = 15 nM for murine JAK1), JAK2 (IC_{50} = 1 nM), JAK3 (Ki = 5 nM), and Tyk2 (IC_{50} = 1 nM).	500 µg 1 mg
InSolution™ JAK Inhibitor I	420097	A 10 mM (500 µg/162 µL) solution of JAK Inhibitor I (Cat. No. 420099) in DMSO.	500 µg
JAK2 Inhibitor II 1,2,3,4,5,6-Hexabromo-cyclohexane	420132	A cell-permeable, specific, reversible, and direct inhibitor of JAK2 autophosphorylation (maximal inhibition at 50 µM in BSC-40 cells overexpressing JAK2).	25 mg
JAK2 Inhibitor III, SD-1029 NSC 371488	573098	A cell-permeable, selective inhibitor of JAK2.	10 mg
JAK2 Inhibitor IV 3-Amino-5-(N-tert-butyl-sulfonamido-4-phenyl)-indazole	420139	A potent inhibitor of both the wild-type JAK2 and the constitutively active V617F mutant (IC_{50} = 78 and 206 nM, respectively).	5 mg
JAK2 Inhibitor IX, WP1193	420151	A cell-permeable analog of WP1130 (Cat. No. 681685) that inhibits JAK2 kinase activity and suppresses JAK2-dependent STAT3 Tyr705 phosphorylation.	10 mg
JAK2 Inhibitor V, Z3 2-Methyl-1-phenyl-4-pyridin-2-yl-2-(2-pyridin-2-ylethyl)butan-1-one	420141	A cell-permeable inhibitor of autophosphorylation of both the wild-type JAK2 and the constitutively active V167F mutant (IC_{50} = 15 and 28 µM, respectively).	10 mg
JAK3 Inhibitor I WHI-P131	420101	A cell-permeable, reversible, potent, ATP-competitive, and specific inhibitor of JAK3 (IC_{50} = 78 µM).	5 mg
JAK3 Inhibitor II WHI-P154	420104	A potent, cell-permeable, reversible, ATP-competitive, and specific inhibitor of JAK3 with no effect on JAK1 and JAK2.	5 mg
JAK3 Inhibitor, Negative Control; WHI-P258, HCl	420112	A useful negative control for JAK3 Inhibitors (IC_{50} > 300 µM).	1 mg
JAK3 Inhibitor IV ZM 39923	420121	A potent, selective, and ATP-competitive inhibitor of Janus tyrosine kinase-3 (JAK3) (pIC_{50} = 7.1).	10 mg
JAK3 Inhibitor V ZM 449829	420122	A breakdown product of JAK3 Inhibitor IV (Cat. No. 420121) with similar inhibitory potency (pIC_{50} = 6.8 for JAK3)	10 mg
JAK3 Inhibitor VI	420126	A cell-permeable, potent, and reversible inhibitor of JAK3 (IC_{50} = 27 nM).	5 mg
JAK3 Inhibitor VII, AD412 N-(Pyridin-4-yl)-3-[1-(4-chloro-benzyl)indol-3-yl]-propanamide	420145	A cell-permeable, selective inhibitor of JAK3 (by 81% and 36% at 90 and 30 µM, respectively)	25 mg
JAK3 Inhibitor VIII NSC114792	420146	A cell-permeable compound that targets JAK3 kinase domain and selectively inhibits JAK3, but not JAK1, JAK2, or TYK2.	10 mg

STAT Inhibitors

Description	Catalogue No.	Comments	Size
STAT3 Inhibitor Peptide Ac-PpYLKTK-OH	573095	A STAT3-SH2 domain binding phosphopeptide that acts as a selective inhibitor of STAT3 signaling (DB_{50} = 235 µM).	1 mg
STAT3 Inhibitor Peptide, Cell-Permeable PpYLKTK-mts	573096	A cell-permeable analog of the STAT3-SH2 domain-binding phosphopeptide (Cat. No. 573095) that contains a C-terminal membrane translocating sequence (MTS).	1 mg 5 mg
STAT3 Inhibitor III, WP1066	573097	A cell-permeable analog of AG 490 (Cat. No. 658401) that acts as a STAT3 pathway inhibitor. Inhibits malignant glioma growth (IC_{50} = 5.6 and 3.7 µM in U87-MG and U373-MG cells, respectively).	10 mg
InSolution™ STAT3 Inhibitor III, WP1066	573129	A 50 mM (5 mg/281 µL) solution of STAT3 Inhibitor III, WP1066 (Cat. No. 573097) in DMSO.	5 mg
STAT3 Inhibitor V, Stattic 6-Nitrobenzo[b]thiophene-1, 1-dioxide	573099	A cell-permeable inhibitor of STAT3 cellular function that targets the STAT3-SH2 domain and prevents its association with upstream kinases.	25 mg

STAT Inhibitors (continued)

Description	Catalogue No.	Comments	Size
STAT3 Inhibitor VI, S3I-201 NSC 74859	573102	A cell-permeable compound that binds STAT3-SH2 domain and prevents STAT3 phosphorylation/activation, dimerization, DNA-binding, and STAT3-dependent transcription.	10 mg 25 mg
InSolution™ STAT3 Inhibitor VI, S3I-201	573130	A 50 mM (5 mg/274 µL) solution of STAT3 Inhibitor VI, S3I-201 (Cat. No. 573102) in DMSO.	5 mg
STAT3 Inhibitor VII Ethyl-1-(4-cyano-2,3,5,6-tetra-fluorophenyl)-6,7,8-trifluoro-4-oxo-1,4-dihydroquinoline-3-carboxylate	573103	A cell-permeable, potent inhibitor of JAK1/JAK2/TYK2/STAT3 activation (IC_{50} = 170 nM against U266 constitutive STAT3 Y705 phosphorylation).	5 mg
STAT3 Inhibitor VIII, 5,15-DPP 5,15-Diphenylporphyrin	573109	A cell-permeable compound that directly binds to STAT3 and prevents its dimerization. Acts as a STAT3-SH2 domain antagonist (IC_{50} = 280 nM).	25 mg
STAT3 Inhibitor X, HJB 17-Hydroxy-jolkinolide B	573106	A highly selective inhibitor of JAK/STAT3 signaling with anti-tumor properties (~12 µM). Suppresses IL-6-induced STAT-3 transcription activity (IC_{50}= ˜ 3.34 µM in HepG2 cells).	1 mg
STAT3 Inhibitor Peptide, Cell-permeable, Inactive Control PYLKTK-mts	573105	A cell-permeable, non-phosphorylated inactive control peptide for STAT3 cellular function studies.	5 mg
STAT3 Inhibitor IX, Cpd188 4-((3-(Carboxymethylsulfanyl)-4-hydroxy-1-naphthyl)sulfamoyl)-benzoic acid	573125	A cell-permeable compound that inhibits IL-6-stimulated STAT3 Tyr705 phosphorylation (IC_{50} = 73 µM) and nuclear translocation (IC_{50} = 39 µM) in HepG2 cells.	10 mg
STAT3 Inhibitor XI, STX-0119 N-(5-(Furan-2-yl)-1,3,4-oxadiazol-2-yl)-2-phenylquinoline-4-carboxamide	573126	A cell-permeable, selective inhibitor of STAT3 DNA binding activity. Prevents IL-6-stimulated STAT3 dimerization in HEK293 cells (IC_{50} = 74 µM).	25 mg
STAT3 Inhibitor XII, SPI H2N-FISKERERAILSTKPP-GTFLLRFSESSK-CO2H	573127	A cell-permeable STAT3-SH2 domain-derived (588-615) 28-mer peptide that competitively prevents STAT3 binding to cognate pTyr peptide motif, represses STAT3 dimerization and induces apoptosis.	5 mg
STAT3 Inhibitor XIII, C188-9 F1113-0789	573128	A cell-permeable binaphthol-sulfonamide that competes against pY proteins for STAT3-SH2 domain binding.	10 mg
STAT3 Inhibitor XIV, LLL12 5-Hydroxy-9,10-dioxo-9,10-dihydroanthracene-1-sulfonamide	573131	A cell-permeable compound that binds directly to the phosphoryl Tyr705 binding site of the STAT3 monomer and acts as selective and reversible inhibitor of STAT3 signaling.	10 mg
STAT3 Inhibitor XVIII, BP-1-102	573132	A cell-permeable compound that selectively binds the three subpockets of STAT3- SH2 domain surface (KD = 504 nM) and blocks STAT3 phosphorylation, dimerization, and DNA-binding.	10 mg
STAT5 Inhibitor II, IQDMA N1-(11H-indolo[3,2-c]quinolin-6-yl)-N2,N2-dimethylethane-1,2-diamine	420294	A cell-permeable inhibitor of STAT5 signaling in HL-60 and K562 cells and inhibits the growth of various tumor cells (IC_{50} = 8, 5, and 8 µM for A549, HL-60, and K562 cells, respectively).	10 mg
STAT5 Inhibitor N′-((4-Oxo-4H-chromen-3-yl) methylene)nicotinohydrazide	573108	A cell-permeable compound that selectively targets the SH2 domain of STAT5 (IC_{50} = 47 µM against STAT5b SH2 domain EPO peptide binding activity).	10 mg
STAT5 Inhibitor III, Pimozide	573110	A cell-permeable compound that inhibits the constitutive STAT5 Tyr694 phosphorylation and transcription activity in Bcr-Abl+ K562 and KU812 cultures.	100 mg
STAT3/STAT5 Dual Inhibitor, SH-4-54	509105	A cell-permeable, salicyclic-to-benzoic acid-substituted BP-1-102 (Cat. No. 573132) analog that targets STAT3 and STAT5 with comparable affinity (k_{off}/k_{on} = K_D = 300 and 464 nM, respectively).	10 mg

JAK/STAT Proteins & Peptides

Description	Applications	Qty/Pk	Catalogue No.
JAK2 Protein, Active	Kinase Assay	10 µg	14-640
JAK2 Protein, immune complex	Kinase Assay	100 µL	14-134
JAK3 Protein, Active	Kinase Assay	10 µg	14-629
JAK3tide	Kinase Assay	1000 µg	12-547

JAK/STAT RNA Detection

Description	Applications	Qty/Pk	Catalogue No.
JAK1, RNA probe, Hu, Cy3	RNA Expression	250 rxns	SF-1288
JAK1, RNA probe, Hu, Cy5	RNA Expression	250 rxns	SF-728
JAK2, RNA probe, Hu, Cy3	RNA Expression	250 rxns	SF-1410
JAK2, RNA probe, Hu, Cy5	RNA Expression	250 rxns	SF-406
JAK3, RNA probe, Hu, Cy3	RNA Expression	250 rxns	SF-905
JAK3, RNA probe, Hu, Cy5	RNA Expression	250 rxns	SF-476

JAK/STAT Kits & Assays

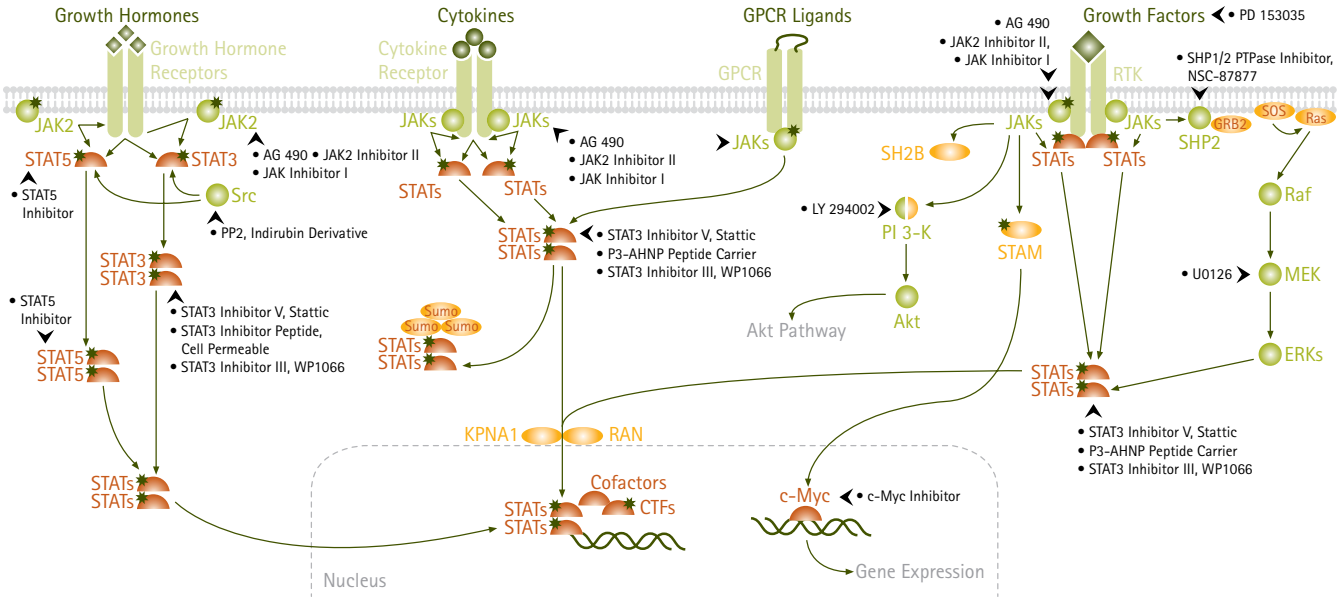
Description	Applications	Qty/Pk	Catalogue No.
STAT1 Activation Dual Detection Kit	Flow Cytometry	25 tests	FCCS025142
Phospho STAT1(Tyr701) Milliplex Assay	Multiplexed ELISA	96 well	46-655MAG
Total STAT1 Milliplex Assay	Multiplexed ELISA	96 well	46-654MAG

InhibitorSelect™ JAK/STAT Signaling Pathway Inhibitor Panel

(Catalogue No. 420138)

The panel contains the following inhibitors and one vial of high quality DMSO.

Description	Qty/Pk	Catalogue No.
PD 153035	1 mg	234490
Indirubin Derivative E804	1 mg	402081
JAK Inhibitor I	500 µg	420099
JAK2 Inhibitor II	25 mg	420132
LY 294002	5 mg	440202
PP2	1 mg	529573
SHP1/2 PTPase Inhibitor, NSC-87877	50 mg	565851
STAT3 Inhibitor Peptide, Cell Permeable	1 mg	573096
STAT3 Inhibitor III, WP1066	10 mg	573097
STAT3 Inhibitor V, Stattic	25 mg	573099
STAT5 Inhibitor	10 mg	573108
AG490	5 mg	658401
U0126	1 mg	662005
DMSO	15 mL	KP31817



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