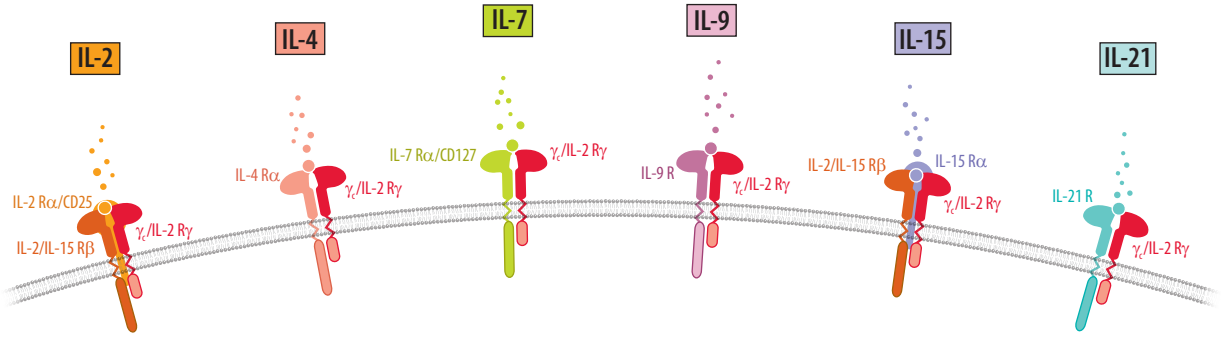
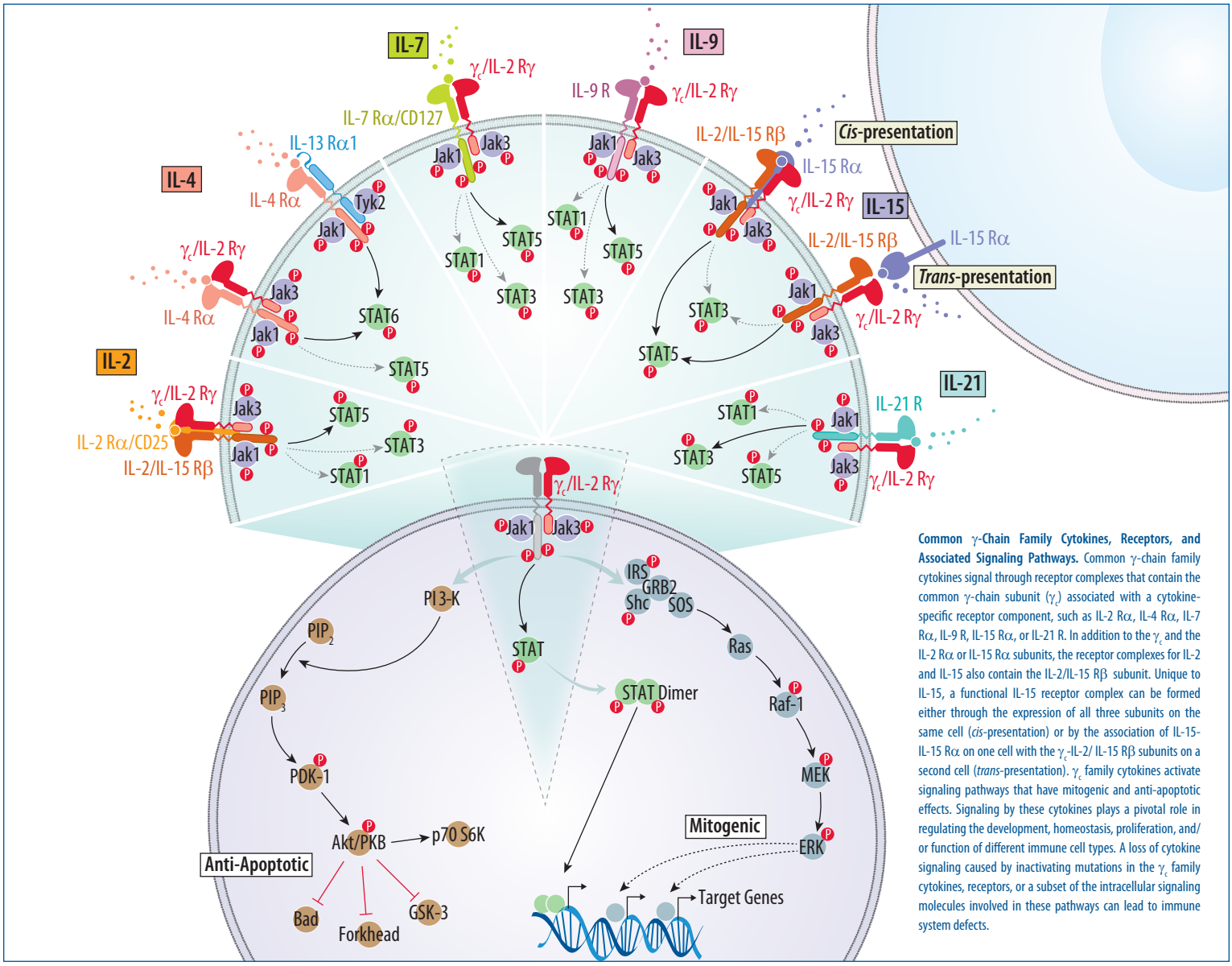
Products for the Common Cytokine Receptor γ-Chain Family



Common γ-Chain Family Cytokines Regulate Immune System Functions

Cytokines belonging to the common cytokine receptor γ-chain (γ_c) family include IL-2, IL-4, IL-7, IL-9, IL-15, and IL-21. Members of this family signal through receptor complexes that contain the γ_c/IL-2 Rγ subunit. The γ_c subunit associates with different cytokine-specific receptor subunits to form unique heterodimeric receptors for IL-4, IL-7, IL-9, and IL-21, or associates with both IL-2/IL-15 Rβ and IL-2 Rα or IL-15 Rα to form heterotrimeric receptors for IL-2 or IL-15, respectively. γ_c family cytokines generally activate three major signaling pathways that promote cellular survival and proliferation, the PI 3-K-Akt pathway, the RAS-MAPK pathway, and the JAK-STAT pathway. Differences in the expression patterns of the cytokines or their unique receptor components, along with the activation of different STAT proteins may account for some of the distinct effects mediated by γ_c family cytokines.

Signaling by γ_c family cytokines plays a major role in regulating the development, survival, proliferation, differentiation and/or function of cells of the immune system. The importance of the γ_c family cytokines for the establishment and maintenance of the immune system is emphasized by the fact that mutations in γ_c/IL-2 Rγ are associated with a disease known as X-linked severe combined immunodeficiency (XSCID), which is characterized by the absence of T cells and natural killer (NK) cells, and the presence of non-functional B cells. Knockout studies in mice have demonstrated that the lack of T cell and NK cell development in this disease can be primarily attributed to the respective loss of IL-7 and IL-15 signaling, while the loss of both IL-4 and IL-21 signaling leads to defective B cell function. Similar studies revealed that in contrast to humans, B cell development in mice also requires IL-7 signaling. Several additional unique and overlapping effects of the γ_c family cytokines on different immune cell types have been documented. A number of these effects are highlighted on the following pages to demonstrate the central role that γ_c family cytokines play in controlling immune system functions. Understanding the mechanisms by which these cytokines act and how their signaling pathways can be regulated may have therapeutic implications not only for a variety of immunodeficient disease states, but also for disorders resulting from aberrant or exaggerated immune system activation. R&D Systems offers a wide range of proteins, antibodies, flow cytometry kits, ELISAs, and activity assays for research related to the common cytokine receptor γ-chain family. For more information, please visit our website at **www.RnDSystems.com/go/CommonGammaChain**.



Common γ-Chain Family Cytokines, Receptors, and Associated Signaling Pathways. Common γ-chain family cytokines signal through receptor complexes that contain the common γ-chain subunit (γ_c) associated with a cytokine-specific receptor component, such as IL-2 Rα, IL-4 Rα, IL-7 Rα, IL-9 R, IL-15 Rα, or IL-21 R. In addition to the γ_c and the IL-2 Rα or IL-15 Rα subunits, the receptor complexes for IL-2 and IL-15 also contain the IL-2/IL-15 Rβ subunit. Unique to IL-15, a functional IL-15 receptor complex can be formed either through the expression of all three subunits on the same cell (*cis*-presentation) or by the association of IL-15-IL-15 Rα on one cell with the γ_c-IL-2/ IL-15 Rβ subunits on a second cell (*trans*-presentation). γ_c family cytokines activate signaling pathways that have mitogenic and anti-apoptotic effects. Signaling by these cytokines plays a pivotal role in regulating the development, homeostasis, proliferation, and/or function of different immune cell types. A loss of cytokine signaling caused by inactivating mutations in the γ_c family cytokines, receptors, or a subset of the intracellular signaling molecules involved in these pathways can lead to immune system defects.

R&D Systems Products for the Common Cytokine Receptor γ-Chain Family

CYTOKINES & RECEPTORS

MOLECULE	RECOMBINANT PROTEINS	ANTIBODIES	ELISAs/FLOW CYTOMETRY KITS
Common γ-Chain/IL-2 Rγ	H M	H M (B/N, FC, WB)	
IL-2	H M R P B Ca CR E F	H M (B/N, ELISA Cap, ELISA Det, FC, IHC, WB) B E (ELISA Cap, ELISA Det, IHC, WB) P (B/N, FC, IHC,WB) R Ca F (B/N, ELISA Cap, ELISA Det, IHC, WB) CR (B/N, WB)	H M R B Ca E F
IL-2 Rα/CD25	H M R Ca	H M (B/N, ELISA Cap, ELISA Det, FC, IHC, WB), R (IHC, WB)	H M H M R
IL-2 Rβ	H	H (B/N, FC, IHC, WB), M (FC, WB)	
IL-4	H M R P B Ca CR E F RM	H P (B/N, ELISA Cap, ELISA Det, FC, IHC, WB) B (IHC, WB) M CR F (B/N, ELISA Cap, ELISA Det, WB), Ca (B/N, WB) R (B/N, ELISA Cap, ELISA Det, IHC, WB) E (ELISA Cap, ELISA Det, IHC, WB)	H M R P CR E F H
IL-4 Rα	H M	H (B/N, FC, IHC, WB), M (FC, WB)	H M
IL-7	H M	H (B/N, ELISA Cap, ELISA Det, IHC, WB), M (B/N, ELISA Cap, ELISA Det, WB)	H M
IL-7 Rα/CD127	H M R	H (FC, WB), M (ELISA Cap, ELISA Det, FC, WB), R (WB)	M
IL-9	H M R	H (B/N, WB), M R (B/N, FC, WB)	
IL-9 R	H R	H (B/N, FC, WB), M R (WB)	
IL-13 Rα1	H M	H (B/N, ELISA Cap, ELISA Det, FC, IHC, WB)	H
IL-15	H M	H (B/N, ELISA Cap, ELISA Det, FC, IHC, WB), M (B/N, ELISA Cap, ELISA Det, IHC, WB)	H M
IL-15 Rα	H M	H (B/N, FC, IHC, WB), M (B/N, FC, WB)	M
IL-21	M Ca	M (B/N, ELISA Cap, ELISA Det, FC, IHC, WB)	M
IL-21 R	H M	H (FC, WB), M (FC, IHC, WB)	

INTRACELLULAR SIGNALING

MOLECULE	RECOMBINANT PROTEINS	ANTIBODIES	ELISAs/ACTIVITY ASSAYS/ExactaChIP
Akt Pan		H M R (FC, IHC, WB)	
Phospho-Akt (T308) Pan			H M R
Phospho-Akt (S473) Pan		H M R (FC, IHC, WB)	H M R
Akt1	H	H (FC, IHC, WB), M R (WB)	H M R
Phospho-Akt1 (S473)			H M
Akt2		H (FC, IHC, WB), M R (IHC, WB)	
Akt3		H (FC, WB)	
Bad		H M (WB)	H
Bcl-2	H	H (B/N, IHC, IP, WB), M (IHC, IP, WB), R (IP, WB)	H
ERK1	H	H M R (IHC, WB)	H
Phospho-ERK1 (T202/Y204)			H M R
ERK1/2		H M R (FC, IHC, WB)	
Phospho-ERK1 (T202/Y204)/ ERK2 (T185/Y187)		H M R (FC, IHC, WB)	H M R
ERK2	H	H M R (IHC, WB)	H M R
Phospho-ERK2 (T185/Y187)			H M R
GRB2		H M R (IHC, WB)	
GSK-3α		H (IHC)	
Phospho-GSK-3α (S21)		H M R (IHC, WB)	H
GSK-3α/β		H M R (FC, WB)	H M R
Phospho-GSK-3α/β (S21/S9)		H M R (FC, IHC, WB)	H M R
GSK-3β	H	H (FC, IHC, WB), M R (FC, WB)	
Phospho-GSK-3β (S9)		H (FC, WB)	H M R
IRS1		H (IHC, WB), M R (WB)	

MOLECULE	RECOMBINANT PROTEINS	ANTIBODIES	ELISAs/ACTIVITY ASSAYS/ExactaChIP
IRS2		H (WB)	
Jak1		H M R (FC, IHC, WB)	
Phospho-Jak1 (Y1022/Y10232)		H (WB)	
Jak3		H (FC, WB)	
Mcl-1		H (IHC, WB)	
MEK1		H M R (WB)	
Phospho-MEK1 (T292)		H (WB)	
Phospho-MEK1 (T386)		H (WB)	
MEK1/MEK2		H M R (IHC, WB)	
Phospho-MEK1 (S218/ S222)/MEK2 (S222/S226)		H M R (IHC, WB)	H M
MEK2	H	H M R (IHC, WB)	
p70 S6 Kinase	H	H M R (FC, IHC, WB)	H M R
Phospho-p70 S6 Kinase (T229)		H (IHC, WB), M R (WB)	
Phospho-p70 S6 Kinase (T389)		H M R (WB)	H M
Phospho-p70 S6 Kinase (T421/S424)		H M R (WB)	H M R
PDK-1	H	H M R (WB)	
PI 3-Kinase p55γ		H M R (WB)	
PI 3-Kinase p85α		H M R (WB)	
PI 3-Kinase p110β		H (WB)	
PI 3-Kinase p110δ		H (WB)	
PI 3-Kinase P110γ		H (WB)	
Raf-1	H	H M R (IHC, WB)	
Phospho-Raf-1 (S301)		H M R X (WB)	

INTRACELLULAR SIGNALING

MOLECULE	RECOMBINANT PROTEINS	ANTIBODIES	ELISAs/ACTIVITY ASSAYS/ExactaChIP
Phospho-Raf-1 (S642)		H M R (WB)	
Ras		H M R (WB)	
SOS2		H (WB)	
STAT1		H (FC, IHC, IP, WB), M (IP, WB)	H M
Phospho-STAT1 (Y701)		H M (FC, WB)	
STAT3		H M R (FC, IHC, IP, WB)	H M
Phospho-STAT3 (Y705)		H (FC, IHC, WB)	H M

MOLECULE	RECOMBINANT PROTEINS	ANTIBODIES	ELISAs/ACTIVITY ASSAYS/ExactaChIP
Phospho-STAT3 (S727)		H (WB)	
STAT5a		H (FC, IHC, IP, WB), M (IHC, IP, WB)	
STAT5a/b		H M (WB)	H M
STAT5b		H M (FC, IP, WB)	
Phospho-STAT5 (Y699)		H M (FC, IHC, WB)	H M
STAT6		H (FC, IHC, IP, WB), M (FC, IP, WB), R (FC, WB)	
Phospho-STAT6 (Y641)		H (FC, WB)	H M

Species Key: H Human M Mouse R Rat B Bovine Ca Canine CR Cotton Rat E Equine F Feline P Porcine RM Rhesus Monkey X *Xenopus*

Application Key: B/N Blocking/Neutralization ELISA Cap ELISA Capture ELISA Det ELISA Detection FC Flow Cytometry IHC Immunohistochemistry IP Immunoprecipitation WB Western blot

MULTIPLEX ARRAYS

Proteome Profiler™ Antibody Arrays

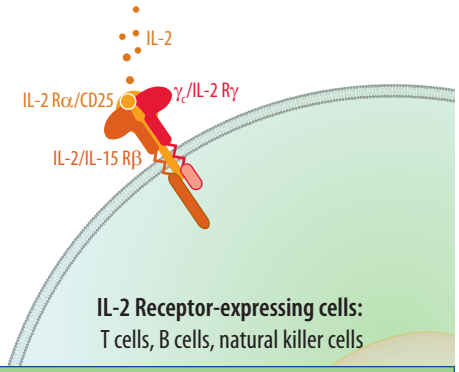
PRODUCT	DESCRIPTION	CATALOG #
Human Apoptosis Array	Contains 4 arrays-each spotted in duplicate with 35 different apoptosis-related antibodies	ARY009
Bad, Bax, Bcl-2, Bcl-x, Pro-Caspase-3, Cleaved Caspase-3, Catalase, Claspin, Clusterin, Cytochrome c, FADD, Fas/TNFSF6, HIF-1α, HO-1/HMOX1/HSP32, HO-2/HMOX2, HSP27, HSP60, HSP70, HTRA2/Omi, cIAP-1, cIAP-2, Livin, p21/CIP1/CDNK1A, p27/Kip1, PON2, Phospho-p53 (S15), Phospho-p53 (S46), Phospho-p53 (S392), Phospho-Rad17 (S635), SMAC/Diablo, Survivin, TNF RI/TNFRSF1A, TRAIL R1/DR4, TRAIL R2/DR5, XIAP		
Human Cytokine Antibody Array, Panel A	Contains 4 arrays-each spotted in duplicated with 36 different cytokine antibodies	ARY005
C5a, CD40 Ligand/CD154, CXCL8/IL-8, G-CSF, GM-CSF, GROα/CXCL1, I-309/CCL1, ICAM-1/CD54, IFN-γ, IL-1α, IL-1β/IL-1F2, IL-1ra, IL-2, IL-4, IL-5, IL-6, IL-10, IL-12 p70, IL-13, IL-16, IL-17, IL-17E, IL-23, IL-27, IL-32α, IP-10/CRG-2/CXCL10, I-TAC/CXCL11, MCP-1/CCL2, MIF, MIP-1α/CCL3, MIP-1β/CCL4, RANTES/CCL5, SDF-1/CXCL12, Serpin E1/PAI-1, TNF-α, TREM-1		
Mouse Cytokine Antibody Array, Panel A	Contains 4 arrays-each spotted in duplicated iwt 40 different cytokine antibodies	ARY006
BLC, C5a, Eotaxin/CCL11, G-CSF, GM-CSF, I-309/CCL1, ICAM-1/CD54, IFN-γ, IL-1α, IL-1β/IL-1F2, IL-1ra, IL-2, IL-3, IL-4, IL-5, IL-6, IL-7, IL-10, IL-12 p70, IL-13, IL-16, IL-17, IL-23, IL-27, IP-10/CRG-2/CXCL10, I-TAC/CXCL11, KC, M-CSF, JE/CCL2/MCP-1, MCP-5/CCL12, MIG/CXCL9, MIP-1α/CCL3, MIP-1β/CCL4, MIP-2/CXCL2/GROβ/CINC-3, RANTES/CCL5, SDF-1/CXCL12, TARC/CCL17, TIMP-1, TNF-α, TREM-1		
Rat Cytokine Antibody Array, Panel A	Contains 4 arrays-each spotted in duplicate with 29 different cytokine antibodies	ARY008
CINC-1/CXCL1, CINC-2α/β/CXCL3, CINC-3/CXCL2, CNTF, Fractalkine/CX3CL1, GM-CSF, ICAM-1/CD54, IFN-γ, IL-1α, IL-1β/IL-1F2, IL-1ra, IL-2, IL-3, IL-4, IL-6, IL-10, IL-13, IL-17, IP-10/CXCL10, LIX, MIG/CXCL9, MIP-1α/CCL3, MIP-3α/CCL20, RANTES/CCL5, L-Selectin/CD62L, Thymus Chemokine/CXCL7, TIMP-1, TNF-α, VEGF		
Human Phospho-Kinase Antibody Array	Contains 4 sets of 2 membranes- each spotted in duplicate with 46 different phospho-specific antibodies	ARY003
Akt (S473), Akt (T308), AMPKα1 (T174), AMPKα2 (T172), β-Catenin, Chk-2 (T68), CREB (S133), ERK1/2 (T202/Y204)/(T185/Y187), FAK (Y397), Fgr (Y412), Fyn (Y420), GSK-3α/β (S21/S9), Hck (Y411), HSP27 (S78/S82), JNK pan (T183/Y185)/(T221/Y223), c-Jun (S63), Lck (Y394), Lyn (Y397), MEK1/2 (S218/S222)/(S222/S226), MSK1/2 (S376/S360), eNOS (S1177), p27 (T157), p27 (T198), p38α (T180/Y182), p53 (S15), p53 (S46), p53 (S392), p70 S6 Kinase (T229), p70 S6 Kinase (T389), p70 S6 Kinase (T421/S424), Paxillin (Y118), PLCγ-1 (Y783), Pyk2 (Y402), RSK1/2 (S221), RSK1/2/3 (S380), Src (Y419), STAT1 (Y701), STAT2 (Y689), STAT3 (Y705), STAT4 (Y693), STAT5a (Y699), STAT5a/b (Y699), STAT5b (Y699), STAT6 (Y641), TOR (S2448), Yes (Y426)		
Human Phospho-MAPK Antibody Array	Contains 4 arrys-each spotted in duplicate with 26 different antibodies recognizing MAPKs and other kinases	ARY002B
Akt1 (S473), Akt2 (S474), Akt3 (S472), Akt pan (S473, S474, S472), CREB, ERK1 (T202/Y204), ERK2 (T185/Y187), GSK-3α/β (S21/S9), GSK-3β (S9), HSP27 (S78/S82), JNK1 (T183/Y185), JNK2 (T183/Y185), JNK3 (T221/Y223), JNK pan (T183/Y185, T221/Y223), MKK3 (S218/T222), MKK6 (S207/T211), MSK2 (S360), p38α (T180/Y182), p38β (T180/Y182), p38δ (T180/Y182), p38γ (T183/Y185), p53 (S46), p70 S6 Kinase (T421/S424), RSK1 (S380), RSK2 (S386), TOR (S2448)		

Proteome Profiler *96* Microplate-based Antibody Arrays

PRODUCT	DESCRIPTION	CATALOG #
Human Phospho-Kinase Array 1	Contains a 96-well microplate spotted with 7 different phospho-kinase antibodies	ARZ004
Akt (S473), ERK1/ERK2 (T202/Y204), GSK-3β (S9), JNK (T183/Y185), p38α (T180/Y182), p70 S6 Kinase (T421/S424), Src (Y416)		

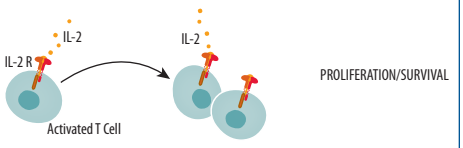
Interleukin-2

Interleukin-2 (IL-2) is an O-glycosylated four α-helix bundle cytokine that is primarily produced by activated T cells, dendritic cells, and B cells.[1,2] It binds with high affinity to a receptor complex that consists of IL-2 Rα/CD25, IL-2 Rβ, and the common γ-chain/IL-2 Rγ subunit. Functionally, IL-2 induces the expression of both IL-2 and IL-2 Rα on activated $CD4^+$ and $CD8^+$ T cells and stimulates their proliferation.[2,3] In contrast, IL-2 also plays an important role in the maintenance of peripheral self-tolerance both by initiating Fas-mediated activation-induced cell death (AICD) of $CD4^+$ T cells following antigen restimulation, and by its ability to promote the development and survival of regulatory T (Treg) cells.[4,5] Rather than displaying a severe immunodeficient phenotype, mice lacking IL-2, IL-2 Rα, or IL-2 Rβ accumulate activated T lymphocytes, have reduced numbers of Treg cells, and develop autoimmune diseases.[6] This suggests that the maintenance of T cell homeostasis and prevention of self-reactivity is the primary function of IL-2 signaling. Other studies demonstrate that IL-2 may enhance the cytotoxicity of natural killer cells and may be required for B cell proliferation and immunoglobulin production.[3,7-10]

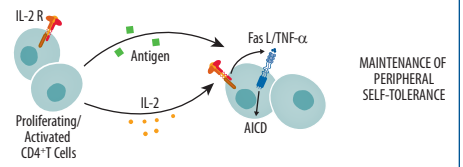


PRIMARY BIOLOGICAL EFFECTS OF IL-2 SIGNALING

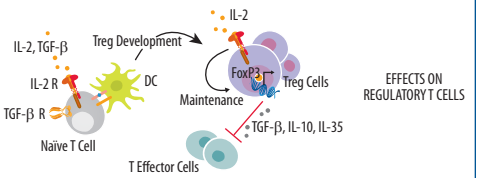
Regulates Activated T Cell Proliferation



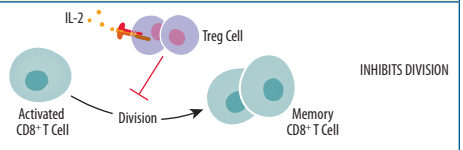
Promotes Activation-induced Cell Death (AICD)



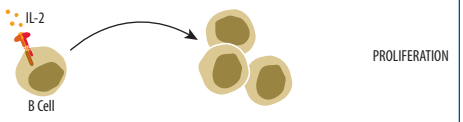
Promotes Treg Cell Development, Survival & Maintenance



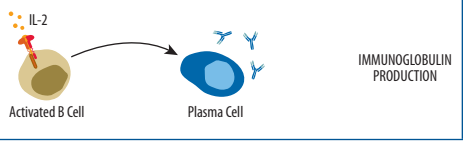
Inhibits Division of $CD8^+$ Memory T Cells



Promotes B Cell Proliferation



Promotes Immunoglobulin Production



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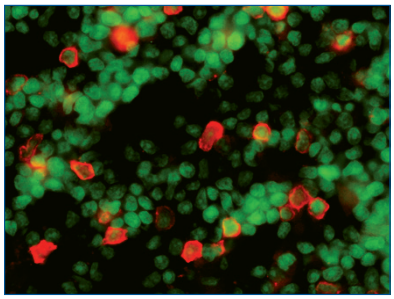
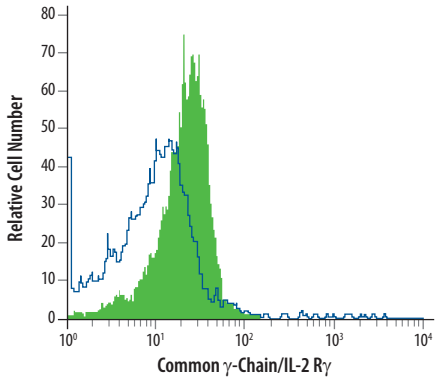
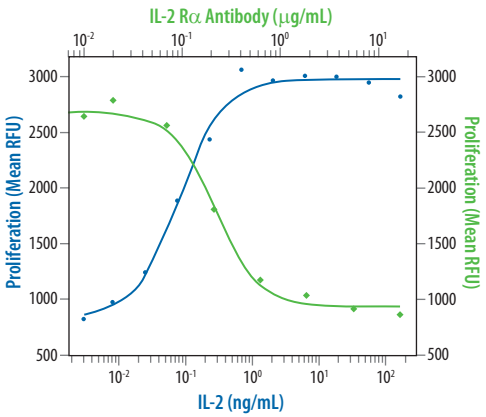


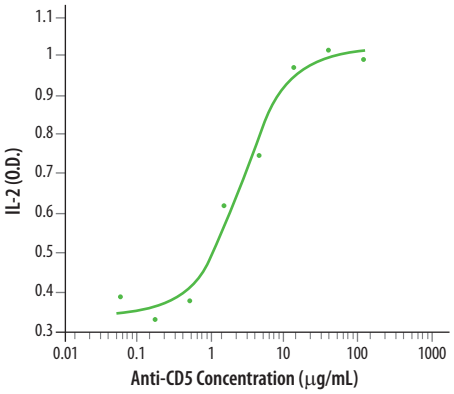
Detection of IL-2 Rα in Mouse Spleen. IL-2 Rα was detected in immersion-fixed frozen sections of mouse spleen using a Goat Anti-Mouse IL-2 Rα Antigen Affinity-purified Polyclonal Antibody (Catalog # AF2438). The tissue was stained using the NorthernLights™ 557-conjugated Donkey Anti-Goat Secondary Antibody (Catalog # NL001; red) and counterstained green.



Detection of Common γ-Chain by Flow Cytometry. Mouse splenic T cells were stained with a PE-conjugated Rat Anti-Mouse Common γ-Chain/IL-2 Rγ Monoclonal Antibody (Catalog # FAB7842P; filled histogram) or a PE-conjugated Rat IgG_{2B} Isotype Control (Catalog # IC013P; open histogram).



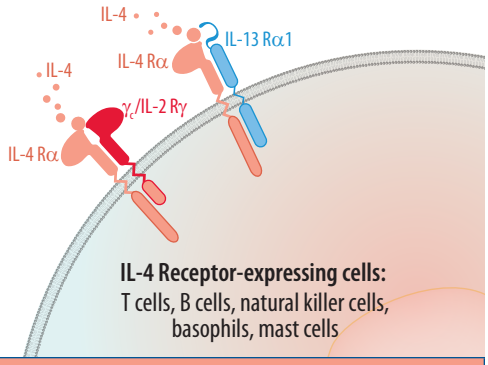
IL-2-induced Proliferation and Neutralization by an Anti-Human IL-2 Rα Antibody. Proliferation of the N1186 human T cell line was assessed following treatment with increasing concentrations of Recombinant Human IL-2 (Catalog # 202-IL; blue line). The stimulatory effect induced by 1 ng/mL Recombinant Human IL-2 was neutralized by treating the cells with increasing concentrations of a Goat Anti-Human IL-2 Rα Antigen Affinity-purified Polyclonal Antibody (Catalog # AF-223-NA; green line).



Measurement of Anti-Human CD5-induced IL-2 Production by Human T Cells. Freshly prepared human T cells were added to a plate coated with suboptimal amounts of a Mouse Anti-Human CD3ε Monoclonal Antibody (Catalog # MAB100) and a Mouse Anti-Human CD28 Monoclonal Antibody (Catalog # MAB342), plus the indicated concentrations of a Goat Anti-Human CD5 Antigen Affinity-purified Polyclonal Antibody (Catalog # AF1636). Following incubation at 37 °C, the levels of IL-2 in the cell culture supernatants were measured using the Human IL-2 Quantikine® ELISA Kit (Catalog # D2050).

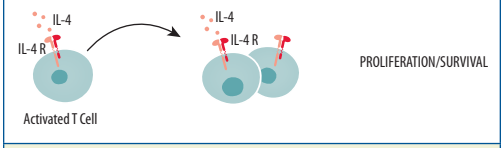
Interleukin-4

Interleukin-4 (IL-4) is a glycosylated, type I cytokine with three intra-chain disulfide bridges that adopts a bundled four α-helix structure.[1] It is primarily produced by T cells, NKT cells, mast cells, and eosinophils.[1,2] IL-4 initiates signal transduction through one of two different receptor complexes, a type I receptor expressed on hematopoietic cells or a type II receptor expressed on nonhematopoietic cells. The type I receptor consists of IL-4 Rα and common γ-chain/IL-2 Rγ and is specific for IL-4, while the type II receptor consists of the IL-4 Rα and IL-13 Rα1 subunits and can be activated by either IL-4 or IL-13. IL-4 signaling is required for the differentiation of Th2 and Th9 cells, and regulates immunoglobulin class switching.[3-7] In addition, IL-4 plays a central role in the development of allergic inflammation and asthma by enhancing the expression of Fcε RI on B cells, mast cells, and basophils, promoting mast cell survival and proliferation, and inducing mast cell, basophil, and eosinophil chemotaxis.[7-12]

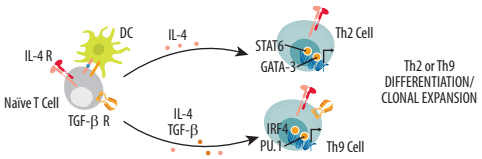


PRIMARY BIOLOGICAL EFFECTS OF IL-4 SIGNALING

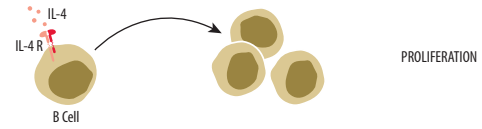
Regulates Activated T Cell Proliferation



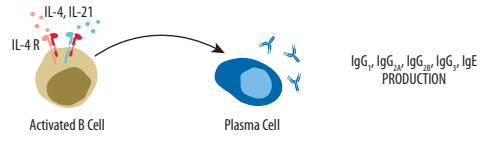
Promotes Th2 or Th9 Differentiation



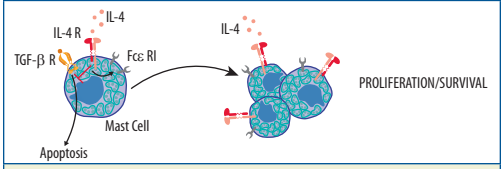
Promotes B Cell Proliferation



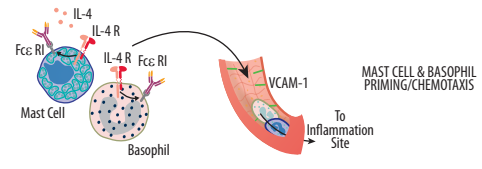
Regulates Immunoglobulin Production & Class Switching



Promotes Mast Cell Survival & Proliferation



Promotes Priming & Chemotaxis of Mast Cells & Basophils



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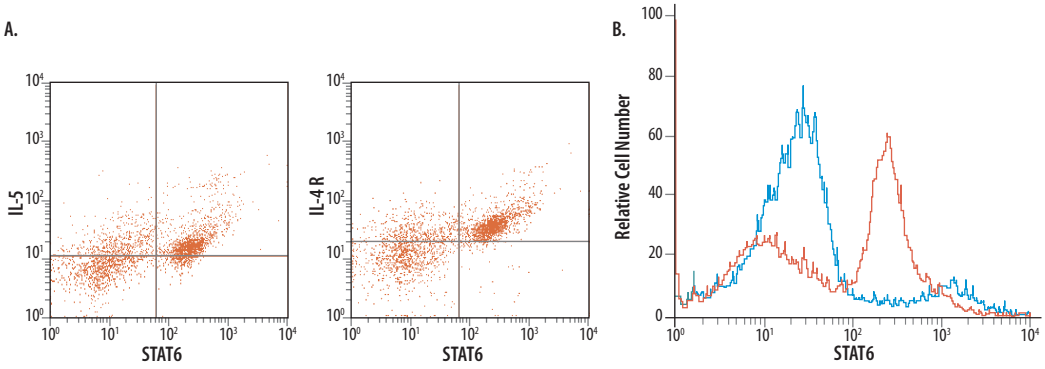
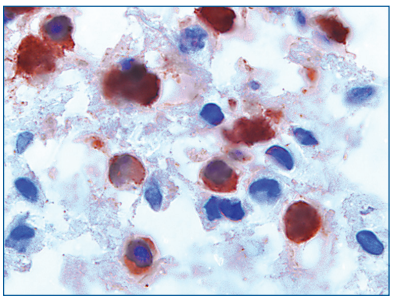
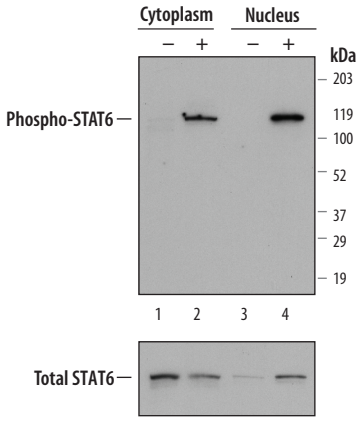


Analysis of Th2 Cell Markers by Multi-color Flow Cytometry. Mouse splenocytes were cultured with Recombinant Mouse IL-4 (Catalog # 404-ML), a Goat Anti-Mouse IFN-γ Antigen Affinity-purified Polyclonal Antibody (Catalog # AF-485-NA), a Hamster Anti-Mouse CD3ε Monoclonal Antibody (Catalog # MAB484), and a Mouse Anti-Human CD28 Monoclonal Antibody (Catalog # MAB342) for 3 days, followed by re-stimulation with PMA/ionomycin to induce Th2 differentiation. **A.** Cells were harvested and stained with the indicated antibodies provided in the Mouse Th2 Cell Multi-Color Flow Cytometry Kit (Catalog # FMC012). Dot plots show the relative IL-4 R^+, IL-5^+, and $STAT6^+$ polulations from activated splenocytes on $CD4^+$-gated cells. Quadrants were set based on isotype controls. **B.** The histogram shows the relative number of $STAT6^+$ cells in unstimulated (blue line) versus stimulated (orange line) splenocytes.



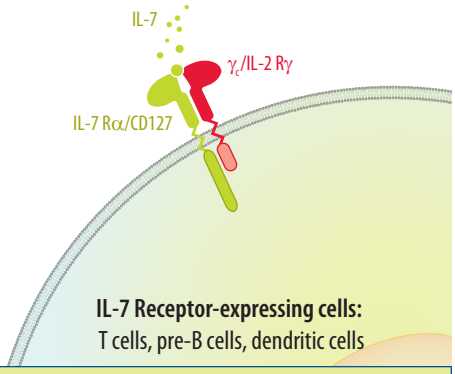
Detection of IL-4 in Human Lymph Node. IL-4 was detected in immersion-fixed frozen sections of human lymph node using a Mouse Anti-Human IL-4 Monoclonal Antibody (Catalog # MAB304). The tissue was stained (red) and counterstained with hematoxylin (blue).



Detection of STAT6 Phosphorylation on Y641 by Western Blot. Cytoplasmic and nuclear extracts from exponentially growing U937 human leukemic monocyte lymphoma cells, untreated (–) or treated (+) with Recombinant Human IL-4 (Catalog # 204-IL), were immunoblotted using a Rabbit Anti-Human STAT6 (Y641) Antigen Affinity-purified Polyclonal Antibody (Catalog # AF3717; top panel). The same membrane was stripped and reprobed with a Mouse Anti-Human STAT6 Monoclonal Antibody (Catalog # MAB2167; bottom panel).

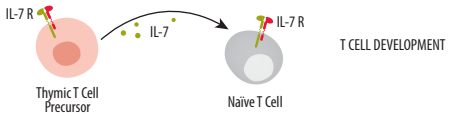
Interleukin-7

Interleukin-7 (IL-7) is a type I glycoprotein that is predicted to form a four α-helix structure with a hydrophobic core.[1] It is produced primarily by stromal cells and exerts its effects through a receptor complex consisting of IL-7 Rα and common γ-chain/IL-2 Rγ. IL-7 signaling is essential for the establishment and maintenance of normal immune system functions.[2] It is required for both mouse and human T cell development and homeostatic proliferation, mouse B cell development, and the generation of $CD4^+$ and $CD8^+$ memory T cells.[2-9] IL-7 Rα-deficient mice have reduced numbers of thymocytes, impaired T cell and B cell development, and lack γδ T cells, a small subset of T cells found in epithelium-rich tissues.[5,6] The requirement of IL-7 for T cell survival has been partially attributed to its ability to induce expression of the anti-apoptotic Bcl-2, Bcl-xL, and Mcl-1 proteins.[10,11] In addition, IL-7 has been found to play a role in regulating V(D)J recombination at the TCRγ, TCRβ, and immunoglobulin heavy chain loci.[12-14]

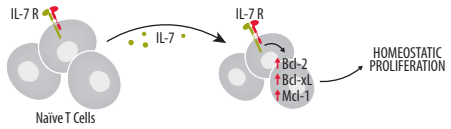


PRIMARY BIOLOGICAL EFFECTS OF IL-7 SIGNALING

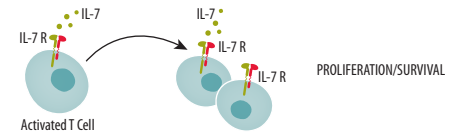
Promotes Human & Mouse T Cell Development



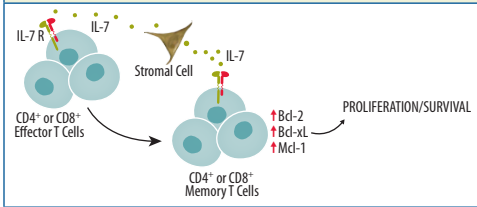
Promotes Naïve T Cell Homeostasis



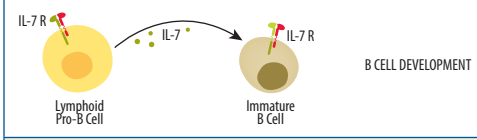
Regulates Activated T Cell Proliferation



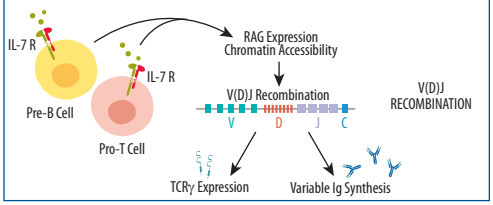
Regulates the Generation & Survival of Memory T Cells



Promotes Mouse B Cell Development



Regulates V(D)J Recombination at the TCR & Distal Ig Heavy Chain Loci



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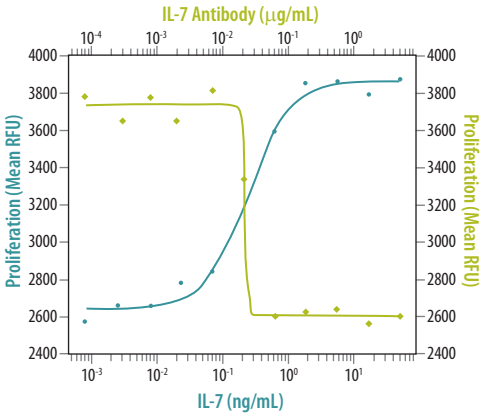
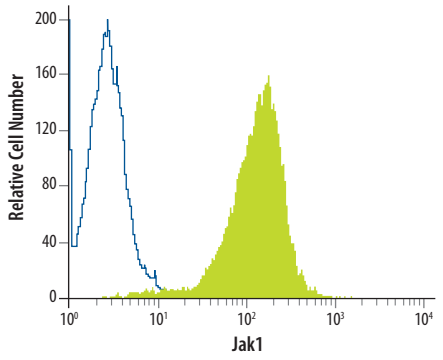
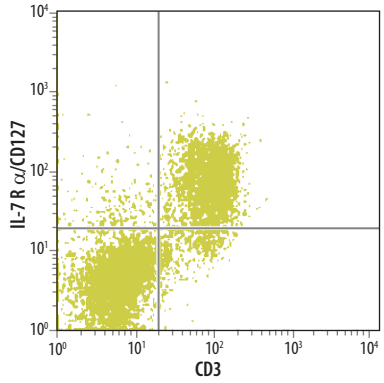


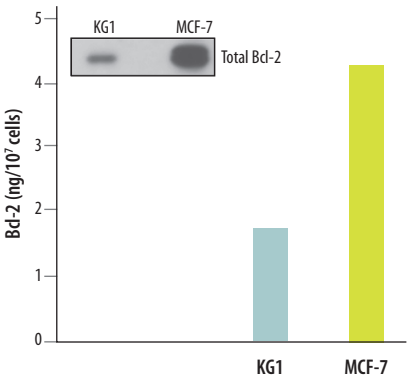
IL-7-induced Proliferation and Neutralization by an Anti-Human IL-7 Antibody. Proliferation of PHA-activated human peripheral blood mononuclear cells was assessed following treatment with increasing concentrations of Recombinant Human IL-7 (Catalog # 207-IL; blue line). The stimulatory effect induced by 2.5 ng/mL Recombinant Human IL-7 was neutralized by treating the cells with increasing concentrations of a Goat Anti-Human IL-7 Antigen Affinity-purified Polyclonal Antibody (Catalog # AF-207-NA; green line).



Intracellular Detection of Jak1 by Flow Cytometry. The Jurkat human leukemic T cell line was stained with a Rat Anti-Human/Mouse/Rat Jak1 Monoclonal Antibody (Catalog # MAB4260; filled histogram) or a Rat IgG_{2B} Isotype Control (Catalog # MAB0061; open histogram), followed by a PE-conjugated Goat Anti-Rat IgG Secondary Antibody (Catalog # F0105B).



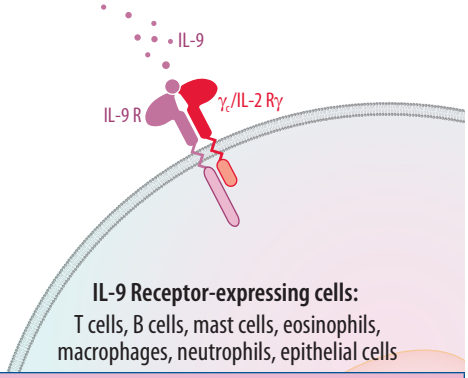
Detection of IL-7 Rα/CD127 by Flow Cytometry. Mouse splenocytes were stained with a PE-conjugated Goat Anti-Mouse IL-7 Rα/CD127 Antigen Affinity-purified Polyclonal Antibody (Catalog # FAB747P). Quadrants were set based on staining with a PE-conjugated Goat IgG Control (Catalog # IC108P).

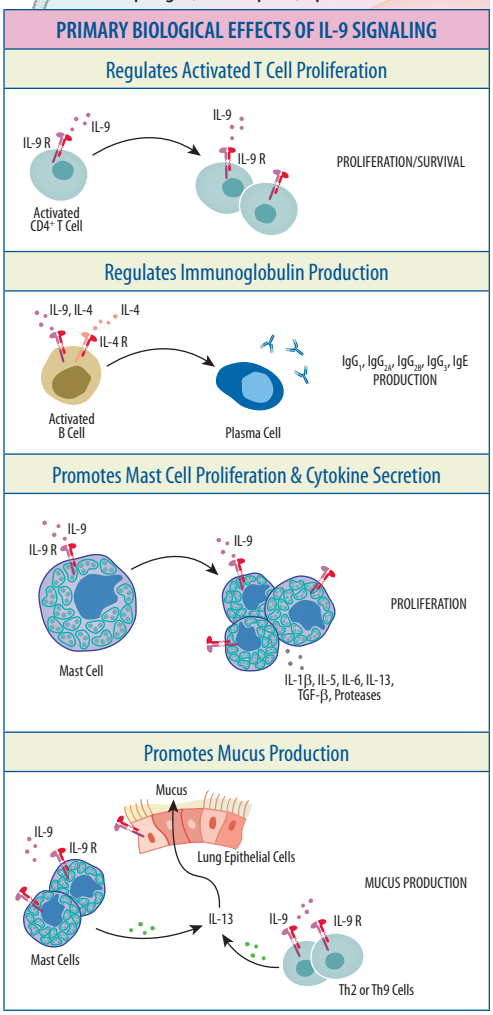


Measurement of Bcl-2 Levels in Cancer Cells using the Bcl-2 DuoSet® IC ELISA. Bcl-2 was measured in cell lysates prepared from KG1 human myeloid leukemia cells and MCF-7 human breast adenocarcinoma cells using the Human Total Bcl-2 DuoSet IC ELISA (Catalog # DYC827B-2; bar graph). The results obtained from the DuoSet IC ELISA are consistent with the relative levels of Bcl-2 detected in the same lysates by Western blot using a Mouse Anti-Human Bcl-2 Monoclonal Antibody (Catalog # MAB827; inset).

Interleukin-9

Interleukin-9 (IL-9) is a pleiotropic cytokine that is produced by activated T lymphocytes.[1] It signals through a receptor complex consisting of IL-9 R and common γ-chain/IL-2 Rγ. IL-9 was initially identified as a mouse T cell and mast cell growth factor.[1-4] Subsequent studies demonstrated that it also regulates immunoglobulin production by B cells, enhances mast cell protease expression, and promotes goblet cell hyperplasia and mucus production, suggesting a link between IL-9 and the development of allergic inflammation.[1,5-7] Although IL-9 was originally thought to be produced primarily by Th2 cells, several studies have recently demonstrated that in the presence of IL-4 and TGF-β, naïve CD4$^+$ T cells differentiate into a distinct IL-9-secreting T cell subset, known as Th9 cells.[8-10] Th9 cells secrete IL-9 and IL-10 (in mice), but do not produce cytokines characteristic of other T helper subsets. Since the precise role of Th9 cells in the pathogenesis of allergic inflammation and other human diseases is currently not well understood, growing interest in this area will help to better define the effects of IL-9 signaling.[11]





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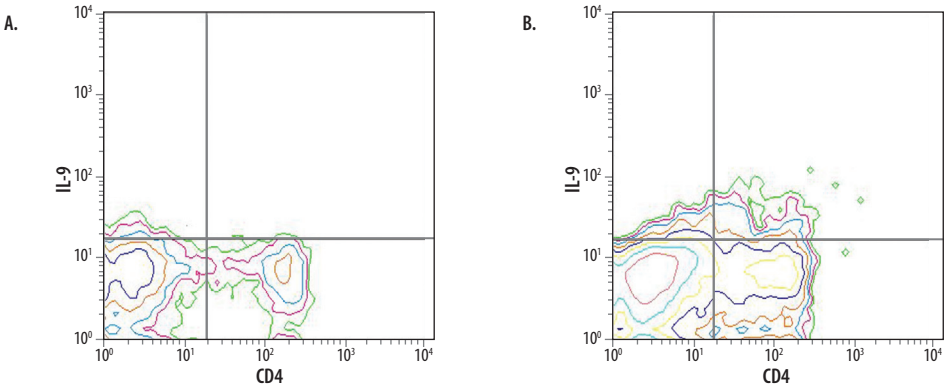
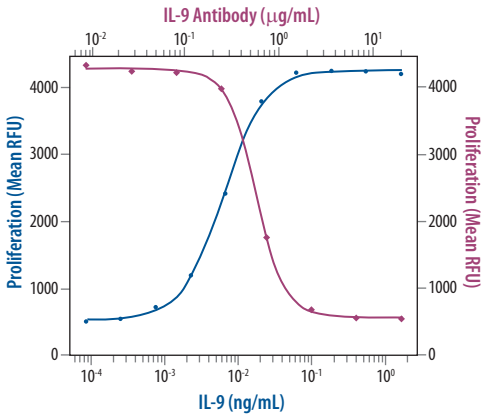
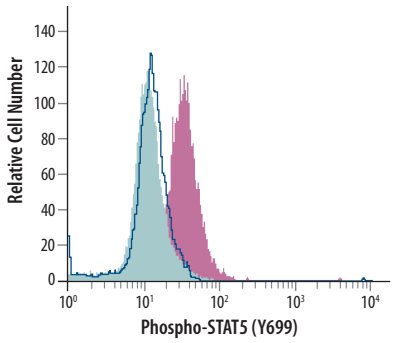


Intracellular Detection of IL-9 by Flow Cytometry. Mouse splenocytes, (**A**) unstimulated or (**B**) stimulated overnight with CD3/CD28/IL-4/TGF-β to induce Th9 development, were stained with a PE-conjugated Rat Anti-Mouse IL-9 Monoclonal Antibody (Catalog # IC409P) and an APC-conjugated Rat Anti-Mouse CD4 Monoclonal Antibody (Catalog # FAB554A). Contour plots were gated on lymphocytes and quadrant markers were set based on isotype controls.



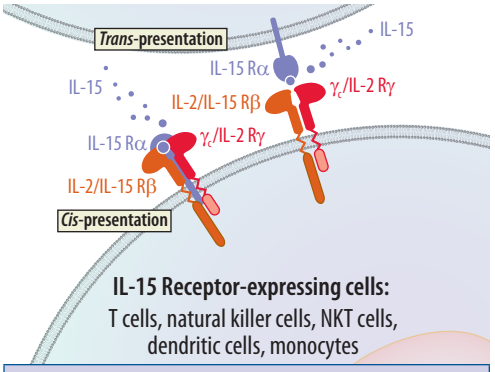
IL-9-induced Proliferation and Neutralization by an Anti-Mouse IL-9 Antibody. Proliferation of the TS1 mouse T helper cell line was assessed following treatment with increasing concentrations of Recombinant Mouse IL-9 (Catalog # 409-ML; blue line). The stimulatory effect induced by 0.05 ng/mL Recombinant Mouse IL-9 was neutralized by treating the cells with increasing concentrations of a Rat Anti-Mouse IL-9 Monoclonal Antibody (Catalog # MAB4091; purple line).



Intracellular Detection of Phospho-STAT5 (Y699) by Flow Cytometry. Untreated (blue filled histogram) or IFN-γ-stimulated (purple filled histogram) Daudi human Burkitt's lymphoma cells were stained with a Rabbit Anti-Human/Mouse Phospho-STAT5 (Y699) Antigen Affinity-purified Polyclonal Antibody (Catalog # AF4190) or a Rabbit IgG Control (Catalog # AB-105-C; open histogram, blue outline), followed by an APC-conjugated Goat Anti-Rabbit IgG Secondary Antibody (Catalog # F0111).

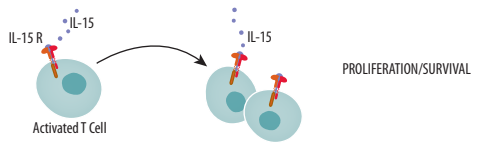
Interleukin-15

Interleukin-15 (IL-15) is a four α-helix bundle cytokine that is structurally and functionally related to IL-2.[1-3] It is produced primarily by dendritic cells, monocytes, and epithelial cells.[4] The heterotrimeric IL-15 receptor consists of a unique IL-15 Rα subunit, IL-2 Rβ, and common γ-chain/IL-2 Rγ. Unlike IL-2, IL-15 binds with high affinity to IL-15 Rα, which then associates with a complex composed of the IL-2 Rβ and common γ-chain/IL-2 Rγ subunits, expressed either on the same cell (*cis*-presentation) or on a different cell (*trans*-presentation).[5] IL-15 signaling is essential for normal immune system functions. It stimulates T cell proliferation and inhibits IL-2-mediated activation-induced cell death (AICD).[1,6,7] In addition, IL-15 is required for the development, survival, and activation of natural killer (NK) cells, homeostasis of natural killer T (NKT) cells and intraepithelial lymphocytes, and maintenance of naïve and memory CD8$^+$ T cells.[8-14] Both IL-15- and IL-15 Rα-deficient mice lack natural killer cells and have severely reduced numbers of NKT cells, memory CD8$^+$ T cells, and specific subsets of intestinal intraepithelial lymphocytes.[15,16]

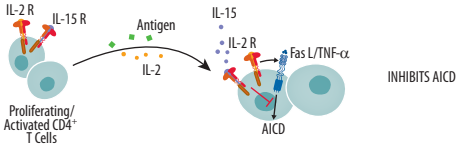


PRIMARY BIOLOGICAL EFFECTS OF IL-15 SIGNALING

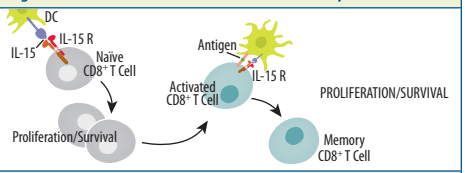
Regulates Activated T Cell Proliferation



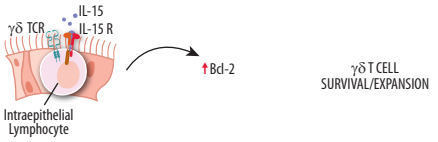
Inhibits Activation-induced Cell Death (AICD)



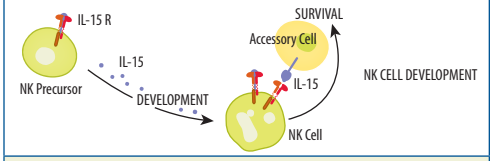
Regulates the Maintenance of Naïve & Memory CD8$^+$ T Cells



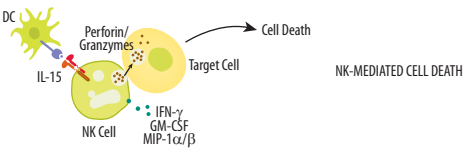
Protects Intestinal Intraepithelial Lymphocytes



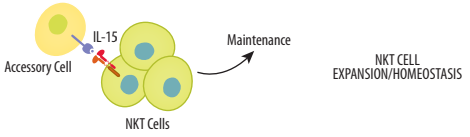
Promotes NK Cell Development & Survival



Promotes Dendritic Cell Priming of NK Cell Activation



Promotes Expansion & Homeostasis of NKT Cells



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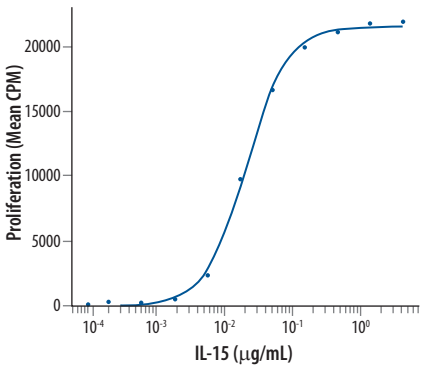
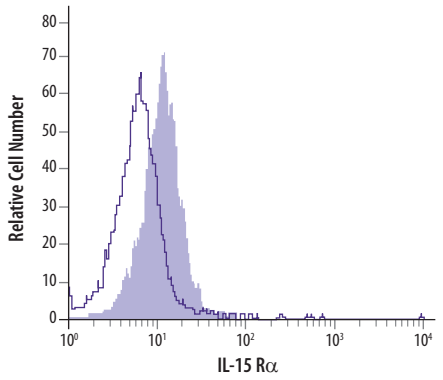
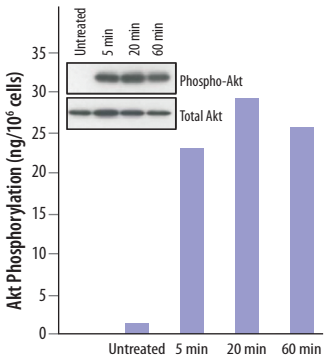


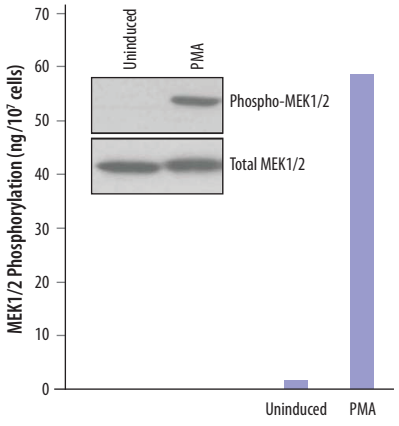
IL-15-induced Proliferation of Mouse Cytotoxic T Cells. The CTLL-2 mouse cytotoxic T cell line was treated with increasing concentrations of Recombinant Mouse IL-15 (Catalog # 447-ML) and proliferation was assessed by measuring ^{3}H-thymidine incorporation.



Detection of IL-15 Rα by Flow Cytometry. The EL-4 mouse lymphoblast cell line was stained with a PE-conjugated Goat Anti-Mouse IL-15 Rα Antigen Affinity-purified Polyclonal Antibody (Catalog # FAB551P; filled histogram) or a PE-conjugated Goat IgG Control (Catalog # IC108P; open histogram).



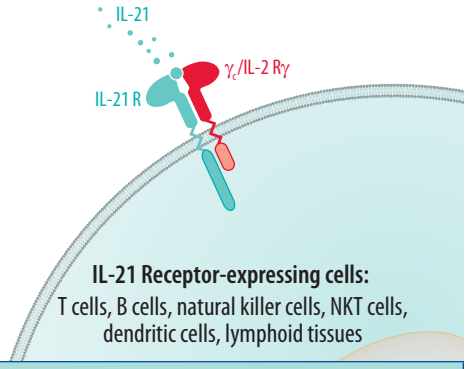
Measurement of IGF-I-induced Akt Phosphorylation on S473 using the Surveyor™ IC Phospho-Akt (S473) ELISA Kit. MCF-7 human breast adenocarcinoma cells were treated with Recombinant Human IGF-I (Catalog # 291-G1) for the indicated times. Akt phosphorylation in cell lysates was assessed using the Human/Mouse/Rat Phospho-Akt (S473) Pan specific Surveyor IC ELISA Kit (Catalog # SUV887; bar graph). The results obtained from the Surveyor IC ELISA were comparable to the relative levels of phosphorylated Akt detected by Western blot (inset).



Quantification of PMA-induced MEK1/MEK2 Phosphorylation using the DuoSet IC ELISA. Lysates from HeLa human cervical epithelial carcinoma cells that were either uninduced or induced with PMA were assessed for MEK1 (S218S222)/MEK2 (S222/S226) phosphorylation using the Human Phospho-MEK1 (S218/S222)/MEK2 (S222/S226) DuoSet IC ELISA (Catalog # DYC2506; bar graph). The results obtained from the DuoSet IC ELISA are consistent with the relative levels of phosphorylated MEK1/2 detected in the same lysates by Western blot.

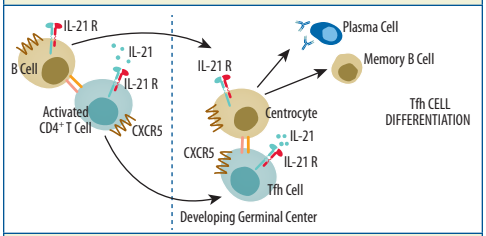
Interleukin-21

Interleukin-21 (IL-21) is the most recently described cytokine belonging to the common cytokine receptor γ-chain family.[1] Like other γ_c family members, IL-21 is a four α-helix bundle type I cytokine that signals through a receptor complex consisting of IL-21 R and common γ-chain/IL-2 Rγ.[2] IL-21 is produced primarily by CD4$^+$ T cells and NKT cells and has a broad range of effects on a number of different cell types.[2,3] IL-21 signaling in CD4$^+$ T cells is required for both Th17 differentiation and the generation of T follicular helper (Tfh) cells, a T cell subset that supports B cell differentiation and antibody production in germinal centers.[3-8] IL-21 also directly regulates B cell proliferation and apoptosis in a context-dependent manner, and can promote immunoglobulin production and isotype class switching.[2,9-11] In addition, IL-21 has been shown to enhance the cytotoxicity of CD8$^+$ T cells, natural killer (NK) cells, and natural killer T (NKT) cells.[9,12-16]

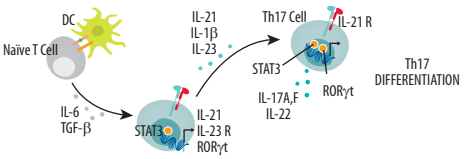


PRIMARY BIOLOGICAL EFFECTS OF IL-21 SIGNALING

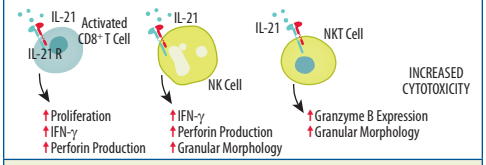
Promotes Tfh Cell Generation & Germinal Center Formation



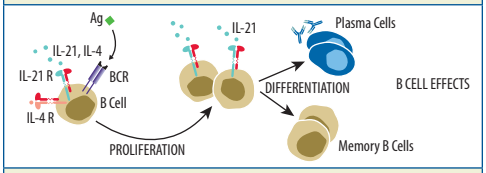
Promotes Th17 Differentiation & Clonal Expansion



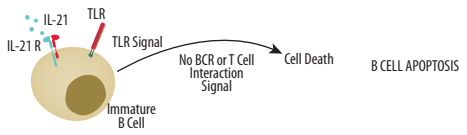
Enhances Cytotoxicity of CD8$^+$ T Cells, NK & NKT Cells



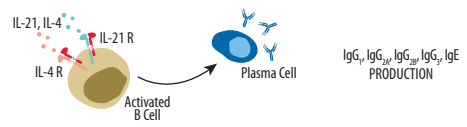
Promotes B Cell Proliferation & Differentiation



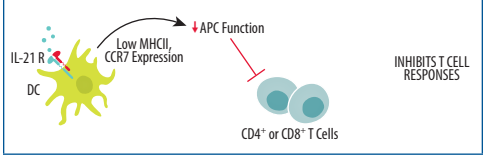
Promotes B Cell Apoptosis in the Absence of BCR & T Cell Signals



Regulates Ig Production & Class Switching



Inhibits Dendritic Cell Maturation & Function



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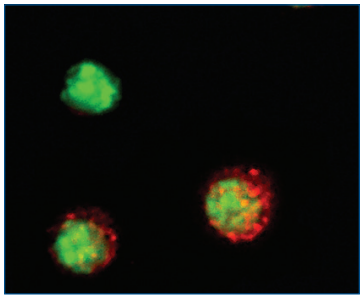
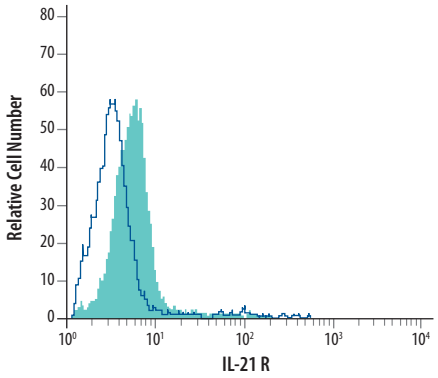
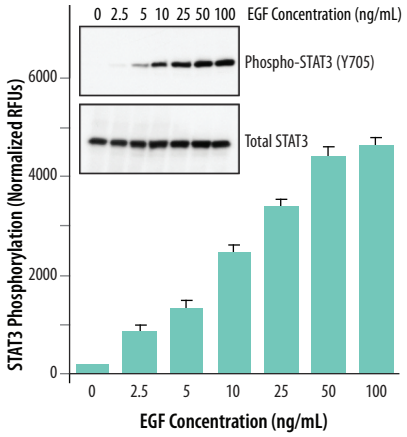


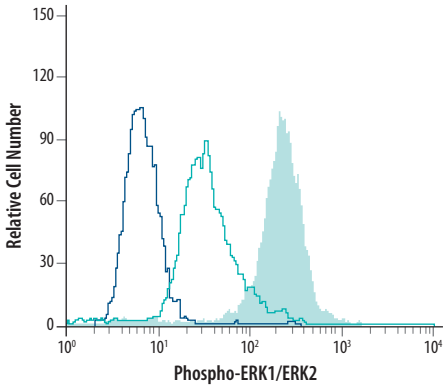
Detection of IL-21 in Mouse Splenocytes. IL-21 was detected in Concanavalin A-stimulated mouse splenocytes using a Goat Anti-Mouse IL-21 Antigen Affinity-purified Polyclonal Antibody (Catalog # AF594) followed by staining with the NorthernLights 557-conjugated Anti-Goat IgG Secondary Antibody (Catalog # NL001; red). Cells were counterstained green.



Detection of IL-21 R by Flow Cytometry. CD19$^+$ human whole blood lymphocytes were stained with a Fluorescein-conjugated Mouse Anti-Human IL-21 R Monoclonal Antibody (Catalog # FAB9911F; filled histogram) or a Fluorescein-conjugated Mouse IgG_1 Isotype Control (Catalog # IC002F; open histogram).



Measurement of EGF-induced STAT3 Phosphorylation on Y705 using the Phospho-STAT3 (Y705) Cell-Based ELISA. A431 human epithelial carcinoma cells were treated with the indicated concentrations of Recombinant Human EGF (Catalog # 236-EG). Phosphorylation of STAT3 on Y705 was determined using the Human/Mouse Phospho-STAT3 (Y705) Cell-Based ELISA (Catalog # KCB4607) and normalized to total STAT3 in the same wells (bar graph). Detection of STAT3 phosphorylation on Y705 by Western blot is shown for comparison (inset).



Intracellular Detection of Phospho-ERK1/2 by Flow Cytometry. The Jurkat human leukemic T cell line, untreated (open histogram, light blue outline) or treated with PMA (filled histogram), was stained with a Rabbit Anti-Human/Mouse/Rat Phospho-ERK1 (T202/Y204)/ERK2 (T185/Y187) Antigen Affinity-purified Polyclonal Antibody (Catalog # AF1018) or a Rabbit IgG Control (Catalog # AB-105-C; open histogram, dark blue outline), followed by a Fluorescein-conjugated Goat Anti-Rabbit IgG Secondary Antibody (Catalog # F0112). To facilitate intracellular staining, the cells were fixed with paraformaldehyde and permeabilized with ice-cold methanol.