#12163 Store at -20°C

IL-10 (D13A11) XP® Rabbit mAb

☐ Small 100 µl
(10 western blots)

☐ Petite 40 µl
(10 western blots)

Cell Signaling TECHNOLOGY®

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New 03/13

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

Entrez-Gene ID #3586
Swiss-Prot Acc. #P22301

Applications	Species Cross-Reactivity*	Molecular Wt.	Isotype
W, F	H	17 kDa	Rabbit IgG**
Endogenous			

Storage: Supplied in 10 mM sodium HEPES (pH 7.5), 150 mM NaCl, 100 µg/ml BSA, 50% glycerol and less than 0.02% sodium azide. Store at –20°C. *Do not aliquot the antibody.*

***Species cross-reactivity is determined by western blot.**

****Anti-rabbit secondary antibodies must be used to detect this antibody.**

Recommended Antibody Dilutions:

Western blotting	1:1000
Flow Cytometry	1:400

For product specific protocols please see the web page for this product at www.cellsignal.com.

Please visit www.cellsignal.com for a complete listing of recommended complementary products.

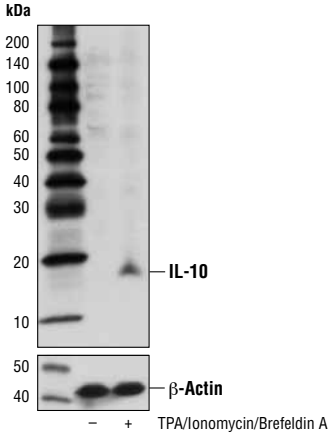
Background: Interleukin-10 (IL-10) is an anti-inflammatory cytokine that is produced by T cells, NK cells, and macrophages (1,2). IL-10 initiates signal transduction by binding to a cell surface receptor complex consisting of IL-10 RI and IL-10 RII (1), leading to the activation of Jak1 and Tyk2 and phosphorylation of Stat3 (1,3). The anti-inflammatory activity of IL-10 is due to its ability to block signaling through other cytokine receptors, notably IFN-γ receptor, by upregulating expression of SOCS1 (1,3). In addition, IL-10 promotes T cell tolerance by inhibiting tyrosine phosphorylation of CD28 (4,5). IL-10 is an important negative regulator of the immune response, which allows for maintenance of pregnancy (1). In contrast, increased IL-10 levels contribute to persistent *Leishmania major* infections (6).

Specificity/Sensitivity: IL-10 (D13A11) Rabbit mAb recognizes endogenous levels of total IL-10 protein.

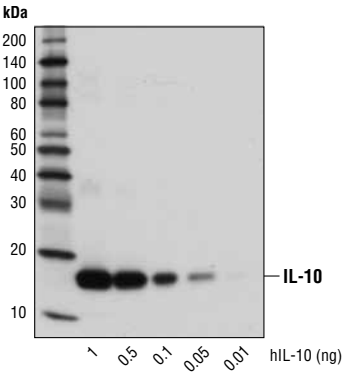
Source/Purification: Monoclonal antibody is produced by immunizing animals with a recombinant human IL-10 protein.

Background References:

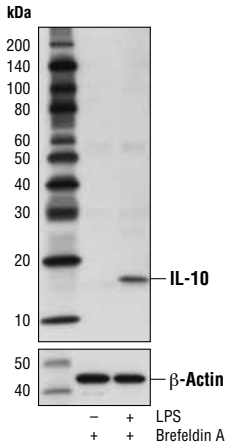
(1) Pestka, S. et al. (2004) *Immunol Rev* 202, 8-32.
(2) Akuffo, H. et al. (1999) *Clin Exp Immunol* 117, 529-34.
(3) O'Shea, J.J. and Murray, P.J. (2008) *Immunity* 28, 477-87.
(4) Akdis, C.A. and Blaser, K. (2001) *Immunology* 103, 131-6.
(5) Akdis, C.A. et al. (2000) *FASEB J* 14, 1666-8.
(6) Von Stebut, E. (2007) Eur J Dermatol 17, 115-22.



Western blot analysis of extracts from purified CD4+ human peripheral blood mononuclear cells, activated with anti-CD3 and anti-CD28 in regulatory T cell inducing conditions for 12 d and then untreated (-) or treated (+) with TPA #4174 (40 nM, 6 hr), Ionomycin #9995 (3 µM, 6 hr), and Brefeldin A #9972 (300 ng/mL, last 5 hr of stimulation), using IL-10 (D13A11) XP® Rabbit mAb (upper) or β-Actin (D6A8) Rabbit mAb #8457 (lower).



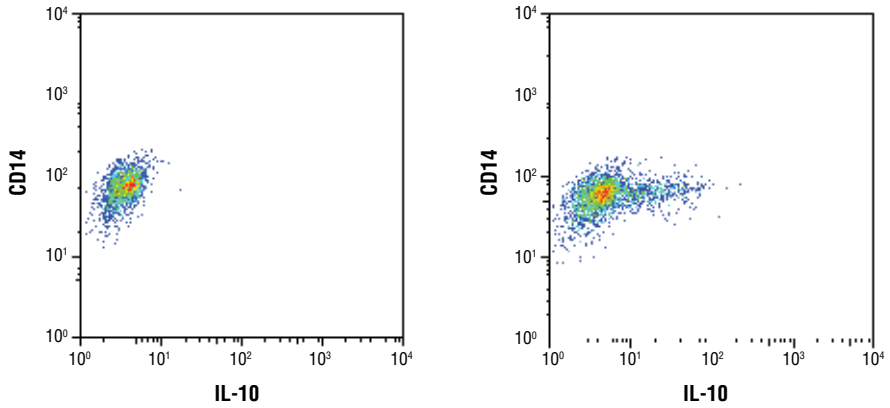
Western blot analysis of recombinant Human Interleukin-10 (hIL-10) #8903 using IL-10 (D13A11) XP® Rabbit mAb.



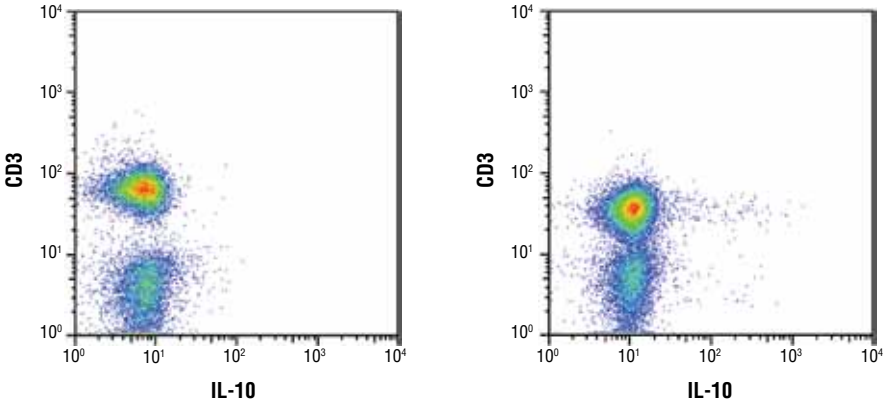
Western blot analysis of extracts from purified CD14+ human peripheral blood mononuclear cells, untreated (-) or treated with LPS (100 ng/mL, 16 hr; +), using IL-10 (D13A11) XP® Rabbit mAb (upper) or β-Actin (D6A8) Rabbit mAb #8457 (lower). Brefeldin A #9972 (300 ng/mL) was added to cells after 1 hr of LPS stimulation.

IMPORTANT: For western blots, incubate membrane with diluted antibody in 5% w/v BSA, 1X TBS, 0.1% Tween-20 at 4°C with gentle shaking, overnight.

Applications Key: **W**—Western **IP**—Immunoprecipitation **IHC**—Immunohistochemistry **ChIP**—Chromatin Immunoprecipitation **IF**—Immunofluorescence **F**—Flow cytometry **E-P**—ELISA-Peptide
Species Cross-Reactivity Key: **H**—human **M**—mouse **R**—rat **Hm**—hamster **Mk**—monkey **Mi**—mink **C**—chicken **Dm**—D. melanogaster **X**—Xenopus **Z**—zebrafish **B**—bovine **Dg**—dog **Pg**—pig **Sc**—S. cerevisiae **Ce**—C. elegans **Hr**—horse **All**—all species expected Species enclosed in parentheses are predicted to react based on 100% homology.



Flow cytometric analysis of purified CD14+ human peripheral blood mononuclear cells untreated (left) or treated with LPS (100 ng/ml, 16 hr; right) using a CD14 antibody and IL-10 (D13A11) XP® Rabbit mAb. Brefeldin A #9972 (300 ng/ml) was added to untreated and treated cells after 1 hr of LPS stimulation. Anti-rabbit IgG (H+L), F(ab')$_2$ Fragment (Alexa Fluor® 647 Conjugate) #4414 was used as a secondary antibody.



Flow cytometric analysis of human peripheral blood mononuclear cells, untreated (left) or treated (right) with TPA #4174 (40 nM, 5 hr), Ionomycin #9995 (2 µM, 5 hr) and Brefeldin A #9972 (1 µg/mL, last 4 hr of stimulation), using a CD3 antibody and IL10 (D13A11) XP® Rabbit mAb. Anti-rabbit IgG (H+L), F(ab')$_2$ Fragment (Alexa Fluor® 647 Conjugate) #4414 was used as a secondary antibody. Analysis was performed on cells in the lymphocyte gate.