Monoclonal Antibodies Detecting Human Antigens

BD FastImmune™ Anti-Human IL-6 (AS12)

 Form
 Catalog Number

 FITC
 340526

 PE
 340527

Product availability varies by region. Contact BD Biosciences Customer Support or your local sales representative for information.

RESEARCH APPLICATIONS

Research applications include studies of:

- Chronic diseases, such as rheumatoid arthritis and multiple sclerosis 1-4
- Neoplasms, such as myelomas and plasmacytomas and Kaposi's sarcoma⁵⁻⁹
- Acute inflammatory disorders such as meningitis, acute hepatitis, and burns^{1,2,10}

DESCRIPTION

Specificity

Antigen distribution

The Anti-Human Interleukin-6 (Anti-Hu–IL-6) antibody recognizes a 21- to 29-kilodalton (kDa) polypeptide. 1,2,11

Human Interleukin-6 is a phosphoglycoprotein with molecular weight ranging from 21 to 29 kDa, depending on the degree of glycosylation and phosphorylation. 1,2,11 IL-6 is a multifunctional cytokine that plays a central role in host defense mechanisms, including acute phase reactions, hematopoiesis, and immune responses. 2,5 Because of its pleiotropic functions, IL-6 has been previously described as interferon- $\beta 2$ (IFN- $\beta 2$), 26K factor, B-cell stimulatory factor 2 (BSF-2), hybridoma/plasmacytoma growth factor (HPGF), hepatocyte-stimulating factor (HSF), and cytotoxic T-cell differentiation factor. 2

IL-6 gene transcription is controlled by at least three factors: activator protein-1 (AP-1) complex, nuclear factor IL-6 (NF IL-6), and nuclear factor kappa B (NF- κ B). ^{2,12,13} IL-6 transcription is downregulated by IL-4 and IL-10. IL-6 transcription is upregulated by bacterial products such as lipopolysaccharide (LPS) or LAM; viruses such as HTLV-1 and HIV; and cytokines such as platelet derived growth factor (PDGF), IL-1, TNF- α , IL-2, and IFN- γ . ^{1,2,5-7,14-16}

The receptor system for IL-6 consists of two polypeptide chains, a low-binding 80-kDa ligand-binding receptor (IL-6R) and a nonbinding 130-kDa signal transducer (gp130). Together the IL-6R and gp130 form a high-affinity complex for IL-6.^{2,17,18}

IL-6 can be expressed by a variety of cells including T lymphocytes, B lymphocytes, monocytes/macrophages, fibroblasts, bone marrow stromal cells, mesangial cells, hepatocytes, keratinocytes, astrocytes, vascular endothelial cells, and various tumor cells. 1,2,5 It induces the final maturation of B lymphocytes into globulin-secreting plasma cells, modulates acute phase proteins in the liver, is an essential accessory factor for T-lymphocyte activation and proliferation, and triggers the emergence of hematopoietic stem cells from the G_0 phase in bone marrow. $^{8,10,19-22}$

For Research Use Only. Not for use in diagnostic or therapeutic procedures.

Becton, Dickinson and Company BD Biosciences 2350 Qume Drive San Jose, CA 95131 USA



Abnormal expression of IL-6 is directly related to the pathogenesis of several diseases, such as multiple myeloma/plasmacytoma and acquired immunodeficiency syndrome (AIDS).^{5-9,12} Elevated quantities of IL-6 are detected in the synovial fluids from affected joints and in sera of patients with active rheumatoid arthritis (RA).^{3,4}

Clone

The Anti-Hu–IL-6 antibody, clone AS12, is derived from the fusion of P3X63.Ag8.653 myeloma cells with splenocytes from BALB/c mice immunized with recombinant human IL-6.

Composition

The Anti-Hu-IL-6 antibody is composed of mouse IgG₁ heavy chains and kappa light chains.

Product configuration

The following are supplied in phosphate buffered saline (PBS) containing a stabilizer and a preservative.

Form	Number of tests	Volume per test (μL) ^a	Amount provided (μg)	Total volume (mL)	Concentration (µg/mL)	Stabilizer	Preservative
FITC	50	20	1.5	1.0	1.5	Gelatin	0.1% Sodium azide
PE	50	20	1.5	1.0	1.5	Gelatin	0.1% Sodium azide

a. Volume required to stain 10⁶ cells.

PROCEDURE

Visit our website (bdbiosciences.com) or contact your local BD representative for the lyse/wash protocol for direct immunofluorescence.

Abbreviated intracellular staining

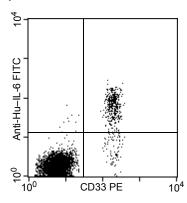
- 1. After surface staining activated whole blood with fluorescent-conjugated monoclonal antibodies, lyse the red blood cells by adding 2 mL of 1X BD FACSTM lysing solution (Cat. No. 349202).
- 2. Vortex gently and incubate 5 to 10 minutes at room temperature.
- 3. Centrifuge at 500 x g for 5 minutes; remove the supernatant.
- 4. Add 500 μL of 1X BD FACSTM Permeabilizing Solution 2 (Cat. No. 347692).
- 5. Vortex and incubate for 10 minutes at room temperature in the dark.
- 6. Wash by adding PBS containing 0.5% bovine serum albumin (BSA) and 0.1% NaN₃, and centrifuge for 5 minutes.
- 7. Add 20 µL of fluorescent-conjugated intracellular antibodies.
- 8. Vortex and incubate for 30 minutes at room temperature in the dark.
- 9. Repeat wash step.
- 10. Resuspend cells in 1% paraformaldehyde in PBS.

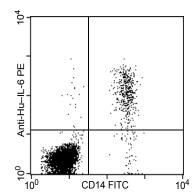
REPRESENTATIVE DATA

Flow cytometric analysis was performed on activated lysed whole blood with a gate set on CD45⁺ mononuclear cells. Laser excitation was at 488 nm.

23-3468-03 Page 2

Figure 1 Four-hour LPS—activated lysed whole blood analyzed with a BD FACS™ brand flow cytometer





HANDLING AND STORAGE

Store vials at 2°C–8°C. Conjugated forms should not be frozen. Protect from exposure to light. Each reagent is stable until the expiration date shown on the bottle label when stored as directed.

WARNING

All biological specimens and materials coming in contact with them are considered biohazards. Handle as if capable of transmitting infection^{23,24} and dispose of with proper precautions in accordance with federal, state, and local regulations. Never pipette by mouth. Wear suitable protective clothing, eyewear, and gloves.

CHARACTERIZATION

To ensure consistently high-quality reagents, each lot of antibody is tested for conformance with characteristics of a standard reagent. Representative flow cytometric data is included in this data sheet.

WARRANTY

Unless otherwise indicated in any applicable BD general conditions of sale for non-US customers, the following warranty applies to the purchase of these products.

THE PRODUCTS SOLD HEREUNDER ARE WARRANTED ONLY TO CONFORM TO THE QUANTITY AND CONTENTS STATED ON THE LABEL OR IN THE PRODUCT LABELING AT THE TIME OF DELIVERY TO THE CUSTOMER, BD DISCLAIMS HEREBY ALL OTHER WARRANTIES, EXPRESSED OR IMPLIED, INCLUDING WARRANTIES OF MERCHANTABILITY AND FITNESS FOR ANY PARTICULAR PURPOSE AND NONINFRINGEMENT. BD'S SOLE LIABILITY IS LIMITED TO EITHER REPLACEMENT OF THE PRODUCTS OR REFUND OF THE PURCHASE PRICE. BD IS NOT LIABLE FOR PROPERTY DAMAGE OR ANY INCIDENTAL OR CONSEQUENTIAL DAMAGES, INCLUDING PERSONAL INJURY, OR ECONOMIC LOSS, CAUSED BY THE PRODUCT.

REFERENCES

- 1. Van Snick J. Interleukin 6: An overview. Ann Rev Immunol. 1990;8:253-278.
- 2. Akira S, Taga T, Kishimoto T. Interleukin-6 in biology and medicine. Adv Immunol. 1993;54:1-79.
- 3. Feldmann M, Brennan FM, Maini RN. Rheumatoid arthritis. Cell. 1996;85:307-310.
- 4. Feldmann M, Brennan FM, Maini RN. Role of cytokines in rheumatoid arthritis. *Ann Rev Immunol*. 1996;14:397-440.
- 5. Kishimoto T. The biology of interleukin-6. Blood. 1989:74:1-10.
- Breen EC, Rezai AR, Nakajuma K, et al. Infection with HIV is associated with elevated IL-6 levels and production. J Immunol. 1990;144:480-484.
- Birx DL, Redfield RR, Tencer K, et al. Induction of interleukin-6 during human immunodeficiency virus infection. Blood. 1990;76:2303-2310.
- 8. Barlogie B, Epstein J, Selvanayagam P, Alexanian R. Plasma cell myeloma: New biological insights and advances in therapy. *Blood*. 1989;73:865-879.
- Van Damme J, Opdenakker G, Simpson RJ, et al. Identification of the human 26-kD protein, interferon β2 (IFN-β₂), as a B cell hybridoma/plasmacytoma growth factor induced by interleukin 1 and tumor necrosis factor. J Exp Med. 1987;165:914-919.

Page 3 23-3468-03

- Gauldie J, Richards C, Harnish D, Lansdorp P, Baumann H. Interferon β2/B-cell stimulatory factor type 2 shares identity with monocyte-derived hepatocyte-stimulating factor and regulates the major acute phase protein response in liver cells. *Proc Natl Acad Sci USA*. 1987;84:7251-7256.
- 11. May LT, Ghrayeb J, Santhanam U, et al. Synthesis and secretion of multiple forms of β2-interferon/B-cell differentiation factor 2/hepatocyte-stimulating factor by human fibroblasts and monocytes. *J Biol Chem.* 1988;263:7760-7765.
- 12. Dokter WHA, Koopmans SB, Vellenga E. Effects of IL-10 and IL-4 on LPS-induced transcription factors (AP-1, NF-IL6 and NF-κB) which are involved in IL-6 regulation. *Leukemia*. 1996;10:1308-1316.
- 13. Libermann TA, Baltimore D. Activation of interleukin-6 gene expression through the NF-κB transcription factor. *Mol Cell Biol.* 1990;10:2327-2334.
- 14. Zhang Y, Broser M, Rom WN. Activation of the interleukin 6 gene by mycobacterium tuberculosis or lipopolysaccharide is mediated by nuclear factors NF-IL6 and NF-κB. Proc Natl Acad Sci USA. 1994;91:2225-2229.
- 15. Iwakura Y, Tosu M, Yoshida E, et al. Induction of inflammatory arthropathy resembling rheumatoid arthritis in mice transgenic for HTLV-1. *Science*. 1991;253:1026-1028.
- Kohase M, May LT, Tamm I, Vilcek J, Sehgal PB. A cytokine network in human diploid fibroblasts: Interactions of β-interferons, tumor necrosis factor, platelet-derived growth factor, and interleukin-1. Mol Cell Biol. 1987;7:273-280.
- 17. Hibi M, Murakami M, Saito M, et al. Molecular cloning and expression of an IL-6 signal transducer, gp130. *Cell.* 1990;63:1149-1157.
- 18. Kishimoto T, Akira S, Taga T. Interleukin-6 and its receptor: A paradigm for cytokines. *Science*. 1992:258:593-597.
- 19. Muraguchi A, Hirano T, Tang B, et al. The essential role of B cell stimulatory factor 2 (BSF-2/IL-6) for the terminal differentiation of B cells. *J Exp Med.* 1988;167:332-344.
- 20. Muraguchi A, Kishimoto T, Miki Y, et al. T-cell replacing factor–(TRF) induced IgG secretion in a human B blastoid cell line and demonstration of acceptors for TRF. *J Immunol.* 1981;127:412-416.
- 21. Lotz M, Jirik F, Kabouridis P, et al. B cell stimulating factor 2/interleukin 6 is a costimulant for human thymocytes and T lymphocytes. *J Exp Med.* 1988;167:1253-1258.
- 22. Koike K, Nakahata T, Takagi M, et al. Synergism of BSF-2/interleukin 6 and interleukin 3 on development of multipotential hemopoietic progenitors in serum-free culture. *J Exp Med.* 1988;168:879-890.
- Protection of Laboratory Workers from Occupationally Acquired Infections; Approved Guideline —
 Third Edition. Wayne, PA: Clinical and Laboratory Standards Institute; 2005. CLSI document M29-A3.
- 24. Centers for Disease Control. Perspectives in disease prevention and health promotion update: universal precautions for prevention of transmission of human immunodeficiency virus, hepatitis B virus, and other bloodborne pathogens in health-care settings. *MMWR*. 1988;37:377-388.

PATENTS AND TRADEMARKS

BD, BD Logo and all other trademarks are property of Becton, Dickinson and Company. © 2014 BD

23-3468-03 Page 4