BD FastImmune™ Anti-Human IL-6 (AS12)

Monoclonal Antibodies
Detecting Human Antigens

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

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 sarcoma\(^5\)\(^-\)\(^9\)
- Acute inflammatory disorders such as meningitis, acute hepatitis, and burns\(^1\)\(^,\)\(^2\)\(^,\)\(^10\)

**DESCRIPTION**

**Specificity**

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

**Antigen distribution**

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-β\(^2\) (IFN-β\(^2\)), 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-kB).\(^2\)\(^,\)\(^12\)\(^,\)\(^13\) 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.\(^8\)\(^,\)\(^10\)\(^,\)\(^19\)\(^-\)\(^22\)
Abnormal expression of IL-6 is directly related to the pathogenesis of several diseases, such as multiple myeloma/plasmacytoma and acquired immunodeficiency syndrome (AIDS). Elevated quantities of IL-6 are detected in the synovial fluids from affected joints and in sera of patients with active rheumatoid arthritis (RA). 

**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.

<table>
<thead>
<tr>
<th>Form</th>
<th>Number of tests</th>
<th>Volume per test (µL)a</th>
<th>Amount provided (µg)</th>
<th>Total volume (µL)</th>
<th>Concentration (µg/µL)</th>
<th>Stabilizer</th>
<th>Preservative</th>
</tr>
</thead>
<tbody>
<tr>
<td>FITC</td>
<td>50</td>
<td>20</td>
<td>1.5</td>
<td>1.0</td>
<td>1.5</td>
<td>Gelatin</td>
<td>0.1% Sodium azide</td>
</tr>
<tr>
<td>PE</td>
<td>50</td>
<td>20</td>
<td>1.5</td>
<td>1.0</td>
<td>1.5</td>
<td>Gelatin</td>
<td>0.1% Sodium azide</td>
</tr>
</tbody>
</table>

a. Volume required to stain 10^6 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.
**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 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.

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**REFERENCES**


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.


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