

## DESCRIPTION

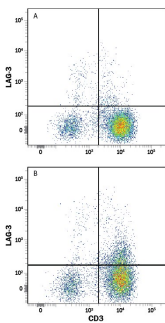
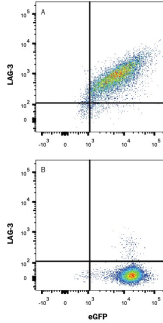
|                           |  |
|---------------------------|--|
| <b>Species Reactivity</b> | Human  |
| <b>Specificity</b>        | Detects human LAG-3 in direct ELISAs.  |
| <b>Source</b>             | Monoclonal Mouse IgG <sub>1</sub> Clone # 874501   |
| <b>Purification</b>       | Protein A or G purified from hybridoma culture supernatant   |
| <b>Immunogen</b>          | Mouse myeloma cell line NS0-derived recombinant human LAG-3<br>Leu23-Leu450<br>Accession # P18627  |
| <b>Conjugate</b>          | Alexa Fluor 700<br>Excitation Wavelength: 675-700 nm<br>Emission Wavelength: 723 nm  |
| <b>Formulation</b>        | Supplied in a saline solution containing BSA and Sodium Azide. See Certificate of Analysis for details.<br><br>*Contains <0.1% Sodium Azide, which is not hazardous at this concentration according to GHS classifications. Refer to the Safety Data Sheet (SDS) for additional information and handling instructions. |

## APPLICATIONS

**Please Note:** Optimal dilutions should be determined by each laboratory for each application. *General Protocols* are available in the *Technical Information* section on our website.

|                       | Recommended Concentration  | Sample    |
|-----------------------|----------------------------|-----------|
| <b>Flow Cytometry</b> | 5 µL/10 <sup>6</sup> cells | See Below |

## DATA

|   |   |
|---|---|
| <p><b>Flow Cytometry</b></p>  <p><b>Detection of LAG-3 in Human PBMCs by Flow Cytometry.</b> Human peripheral blood mononuclear cells (PBMCs) either (A) untreated or (B) treated with 1 µg/mL PHA for 5 days were stained with Mouse Anti-Human LAG-3 Alexa Fluor® 700-conjugated Monoclonal Antibody (Catalog # FAB23193N) and Mouse Anti-Human CD3ε PE-conjugated Monoclonal Antibody (Catalog # FAB100P). Quadrant markers were set based on control antibody staining (Catalog # IC002N). View our protocol for <a href="#">Staining Membrane-associated Proteins</a>.</p> | <p><b>Flow Cytometry</b></p>  <p><b>Detection of LAG-3 in HEK293 Human Cell Line Transfected with Human LAG-3 and eGFP by Flow Cytometry.</b> HEK293 human embryonic kidney cell line transfected with either (A) human LAG-3 or (B) irrelevant transfectants and eGFP was stained with Mouse Anti-Human LAG-3 Alexa Fluor® 700-conjugated Monoclonal Antibody (Catalog # FAB23193N). Quadrant markers were set based on control antibody staining (Catalog # IC002N, data not shown). View our protocol for <a href="#">Staining Membrane-associated Proteins</a>.</p> |
|---|---|

## PREPARATION AND STORAGE

|                                |  |
|--------------------------------|--|
| <b>Shipping</b>                | The product is shipped with polar packs. Upon receipt, store it immediately at the temperature recommended below.                                    |
| <b>Stability &amp; Storage</b> | <b>Protect from light. Do not freeze.</b> <ul style="list-style-type: none"> <li>● 12 months from date of receipt, 2 to 8 °C as supplied.</li> </ul> |

**BACKGROUND**

LAG-3 (Lymphocyte activation gene-3), also known as CD223, is a member of the immunoglobulin superfamily (IgSF). The mature LAG-3 protein is a 496 amino acid (aa) membrane protein with a 421 aa extracellular region which contains four IgSF domains, a 21 aa transmembrane region and a 54 aa cytoplasmic region. LAG-3 and CD4 molecules share < 20% aa sequence homology but have a similar structure (1, 2). Both molecules bind to MHC class II. LAG-3 binds to MHC class II with higher affinity compared to CD4. Both LAG-3 and CD4 genes are located on the distal part of the short arm of chromosome 12.

LAG-3 is an activation-induced molecule, expressed on activated T cells and NK cells, but not on resting T cells. Studies using LAG-3<sup>-/-</sup> mice have shown significant delay of T cell apoptosis following antigen stimulation and increased size of memory T cells pool following infection (3, 4). It also has been reported that anti-LAG-3 antibodies up-regulate T cell activation by blocking interaction of LAG-3 and MHC class II. The study has demonstrated that LAG-3 is selectively expressed on activated CD4<sup>+</sup>CD25<sup>+</sup> T<sub>Reg</sub> cells and plays a role in their suppressive activity (5). This evidence indicated, unlike the interaction of CD4 with MHC class II that plays a positive role in T cell activation, LAG-3 binds to MHC class II and negatively regulates T cell activation through LAG-3 signaling. On the other hand, studies have shown that binding of LAG-3 to MHC class II molecules on antigen presenting cells induce maturation of dendritic cells and cytokine secretion by monocytes through MHC class II signal transduction (6). Taken together, LAG-3 may have two major functions, it negatively regulates T cells activation through LAG-3 signaling and stimulates antigen presenting cells which express MHC class II.

**References:**

1. Triebel, F. *et al.* (1990) J. Exp. Med. **171**:1393.
2. Baixeras, E. *et al.* (1992) J. Exp. Med **176**:327.
3. Workman, C.J. and D.A. Vignali (2003) Eur. J. Immunol. **33**:970.
4. Workman, C.J. *et al.* (2004) J. Immunol. **172**:5450.
5. Huang, C.T. *et al.* (2004) Immunity **21**:503.
6. Andraea, S. *et al.* (2003) Blood **102**:2130.

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