

DESCRIPTION

Species Reactivity	Human
Specificity	Detects human LAG-3 in direct ELISAs and Western blots. In direct ELISAs and Western blots, less than 1% cross-reactivity with recombinant mouse LAG-3 is observed.
Source	Polyclonal Goat IgG
Purification	Antigen Affinity-purified
Immunogen	Mouse myeloma cell line NS0-derived recombinant human LAG-3 Leu23-Leu450 Accession # P18627
Conjugate	Phycoerythrin Excitation Wavelength: 488 nm Emission Wavelength: 565-605 nm
Formulation	Supplied in a saline solution containing BSA and Sodium Azide. See Certificate of Analysis for details. *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
Flow Cytometry	10 μ L/10 ⁶ cells	See Below

DATA

Flow Cytometry

Detection of LAG-3 in Human PBMCs by Flow Cytometry. CD3+ PHA-treated peripheral blood mononuclear cells (PBMCs) were stained with Mouse Anti-Human CD3 ϵ APC-conjugated Monoclonal Antibody (Catalog # [FAB100A](#)) and either (A) Goat Anti-Human LAG-3 PE-conjugated Antigen Affinity-purified Polyclonal Antibody (Catalog # [FAB2319P](#)) or (B) Normal Goat IgG Phycoerythrin Control (Catalog # [IC108P](#)). View our protocol for [Staining Membrane-associated Proteins](#).

Flow Cytometry

Detection of LAG-3 in HEK293 Human Cell Line Transfected with Human LAG-3 and eGFP by Flow Cytometry. HEK293 human embryonic kidney cell line transfected with either (A) human LAG-3 or (B) irrelevant transfectants and eGFP was stained with Goat Anti-Human LAG-3 PE-conjugated Antigen Affinity-purified Polyclonal Antibody (Catalog # [FAB2319P](#)). Quadrant markers were set based on control antibody staining (Catalog # [IC108P](#), data not shown). View our protocol for [Staining Membrane-associated Proteins](#).

PREPARATION AND STORAGE

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

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^{-/-} 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⁺CD25⁺ T_{Reg} 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. Andrae, S. *et al.* (2003) *Blood* **102**:2130.