Accurus Biosciences

Human LAG-3 CHO Stable Cells

Catalog Number:	C1010
Cell Line Name:	CHO suspension cell pool expressing full-length human LAG-3 receptor
Gene name:	LAG-3, human
Gene Sequence:	Codon optimized from NP_002277.4 (Met1-Leu525)
Host Cell:	Suspension CHO
Quantity:	Two vials of frozen cells ($2x10^7$ per vial)
Lot Number:	12042
Stability:	>10 passages
Freeze Media:	Culture media with 10% DMSO
Storage:	liquid nitrogen immediately upon receipt
Culture Medium:	50% CD-CHO (Gibco#10743-029), 50% Ex-Cell CHO 5 Media (Sigma#C0363), supplemented with 8mM L-Glutamine, 1xHT, 1x Penn-Strep and 20ug/ml puromycin.
Mycoplasma Test:	Negative
Application:	Antibody binding assays, IHC/Western blot analysis, or use as cell immunogen

I. Background:

Lymphocyte-activation protein 3 (LAG-3) belongs to the immunoglobulin superfamily and shares sequence homology, exon/intron organization and chromosomal localization to CD4. LAG-3 cDNA encodes a 498 amino acid transmembrane protein with 4 extracellular Ig-like domains. The expression of LAG-3 is undetectable in resting peripheral blood lymphocytes but it is induced in activated T and NK cells. Human MHC Class II molecules can bind to LAG-3 and are considered as receptors for LAG-3 on B cells and dendritic cells. LAG-3 is proven to be an inhibitory receptor on activated T cells. Crosslinking of LAG-3 on activated T cells with an anti-LAG-3 antibody inhibits T cell proliferation and cytokine secretion. Anti-LAG-3 antibodies which blocked LAG-3 binding to MHC class II molecules had strong anti-tumor activity when used in combination with anti-PD-1 antibodies in tumor xerograph models.

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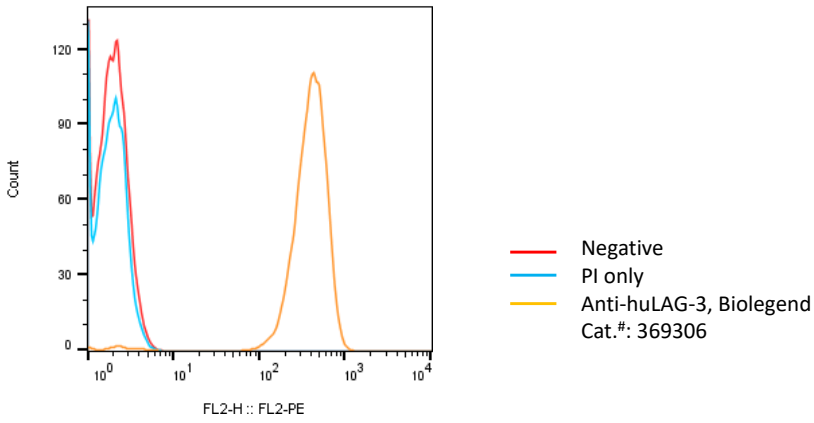
II. Representative Data:

Detection of human LAG-3 expression on human LAG-3 CHO Stable cells using a mouse monoclonal antibody specific for human LAG-3.



III. Thawing and Subculturing:

1. Remove the cell vial from liquid nitrogen tank and thaw cells quickly in a 37°C water bath.
2. In a T250 shake flask, add 30ml cell culture media which has been pre-warmed in a 37°C water bath for 30 minutes.
3. Transfer all the cells in the vial into the T250 shake flask and place it on a shaker inside of a CO2 incubator with 5-8% CO2. The shaker speed should be set at 120-150rpm.
4. The next day, count the cells using a hemocytometer. The viable cell density should be around 0.5-1x10^6/ml and the cell viability should be more than 50%.
5. The cell doubling time should be 18-24 hours. Split the cells 1:5 or 1:10 every 2-3 days or when the cell density reached 6-8x10^6/ml.

IV. References:

Triebel F. *et al.*, LAG-3, a novel lymphocyte activation gene closely related to CD4. *J Exp Med*, 1990 May 1; 171(5): 1393-405.

Baixeras E. *et al.*, Characterization of the lymphocyte activation gene 3-encoded protein. A new ligand for human leukocyte antigen class II antigens. *J Exp Med*, 1992 Aug 1;176(2):327-37.

Hannier E., et al., CD3/TCR complex-associated lymphocyte activation gene-3 molecules inhibit CD3/TCR signaling. J Immunol. 1998 Oct 15;161(8):4058-65.

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