

Human LAG-3 (Luc) Jurkat Reporter Cell Development Service Data Sheet

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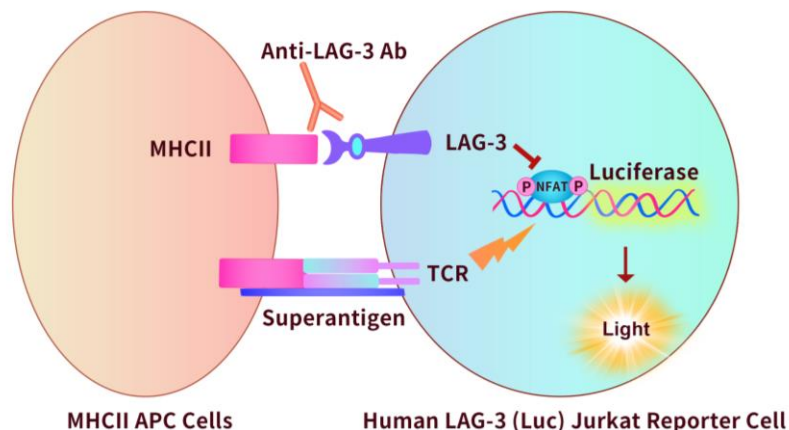
Catalog No.	Size
SCJUR-STF065	2 × (1 vial contains ~5×10 ⁶ cells)

• Description

The Human LAG-3 (Luc) Jurkat Reporter Cell was engineered to not only express the NFAT response element driving luciferase expressing systems, but also express the receptor full length human LAG-3 (Gene ID: 3902), which can use to evaluate the potency of LAG-3 blockade. When co-cultured with target cells expressing human MHCII, the LAG-3/MHCII interaction inhibits TCR signaling and NFAT-mediated luminescence. Blocking the LAG-3/MHCII interaction by anti-LAG-3 antibodies releases the inhibitory signals and results in TCR activation and NFAT-mediated luminescence.

• Application

- Screen for anti-human LAG-3 antibody.



• Cell Line Profile

Cell line	Human LAG-3 (Luc) Jurkat Reporter Cell
Host Cell	Jurkat
Property	Suspension
Complete Growth Medium	RPMI-1640 + 10% FBS
Selection Marker	Hygromycin (20 µg/mL) + Puromycin (5 µg/mL)
Incubation	37°C with 5% CO ₂
Doubling Time	16-20 hours
Transduction Technique	Lentivirus

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• *Materials Required for Cell Culture*

- RPMI Medium 1640 (Gibco, Cat.No.11875-093)
- Fetal bovine serum (CellMax, Cat.No.SA211.02)
- Puromycin (InvivoGen, Cat.No.ant-pr-5b)
- Hygromycin B (Invitrogen, Cat.No.10687010)
- Complete Growth Medium: RPMI-1640 + 10% FBS
- Culture Medium: RPMI-1640 + 10% FBS, Hygromycin (20 µg/mL), Puromycin (5 µg/mL)
- Freeze Medium: 90% FBS, 10% (V/V) DMSO
- T-75 Culture flask (Corning, 430641)
- Cryogenic storage vials (SARSTEDT, 72.379.007)
- Thermostat water bath
- Centrifuge
- Luna cell counter (Logos Biosystems, LUNA- II)
- CO₂ Incubator (Thermo, 3111)
- Biological Safety Cabinet (Thermo, 1389)

• *Recovery*

1. Thaw the vial by gentle agitation in a 37°C water bath. To reduce the possibility of contamination, keep the cap out of the water. Thawing should be rapid (approximately 5 minutes).
2. Remove the vial from the water bath as soon as the contents are thawed, and decontaminate by spraying with 70% ethanol. All the operations from this point on should be carried out under strict aseptic conditions.
3. Transfer the vial contents to a centrifuge tube containing 4.0 mL complete growth medium.
4. Count viable cells and spin at approximately 1000 rpm for 5 minutes.
5. Discard the supernatant and resuspend the cell pellet in an appropriate amount of fresh complete growth medium. Adjust the cell density of the suspension to 1×10⁶ viable cells/mL and transfer cells to an appropriate size vessel.
6. Incubate at 37°C with 5% CO₂ incubator.

Human LAG-3 (Luc) Jurkat Reporter Cell Development Service Data Sheet

• *Subculture*

Adjust the cell density at 2×10^5 - 5×10^5 viable cells/mL by the addition of fresh culture medium or replacement of culture medium. Do not allow the cell density to exceed 3×10^6 cells/mL. T-75 flasks are recommended for subculturing.

- **Medium Renewal:** Add fresh culture medium every 3 to 4 days (depending on cell density)

• *Cryopreservation*

1. Count viable cells and harvest the cell suspension.
2. Centrifuge at 1000 rpm for 5 min at RT and resuspend cells in freezing medium to a concentration of 5×10^6 to 1×10^7 cells/mL.
3. Aliquot into cryogenic storage vials. Place vials in a programmable cooler or an insulated box placed in a -80°C freezer overnight, then transferring to liquid nitrogen storage.

• *Storage*

- **Product format:** Frozen
- **Storage conditions:** Liquid nitrogen immediately upon receipt

Human LAG-3 (Luc) Jurkat Reporter Cell Development Service Data Sheet

• *Receptor Assay*

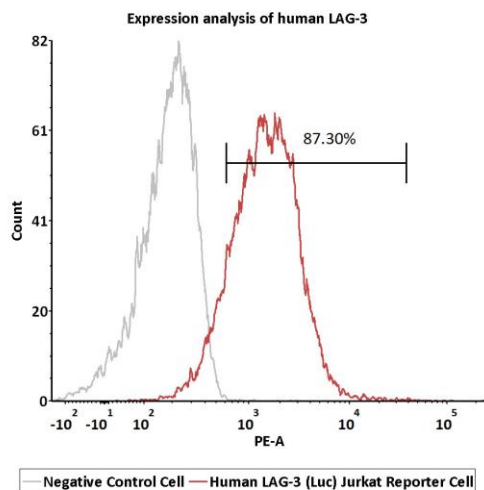


Fig1. Expression analysis of human LAG-3 on Human LAG-3 (Luc) Jurkat Reporter Cell by FACS. Human LAG-3 (Luc) Jurkat Reporter Cell or negative control cell were stained with PE-labeled anti-human LAG-3 antibody.

• *Application*

Anti-human LAG-3 Antibody Screening

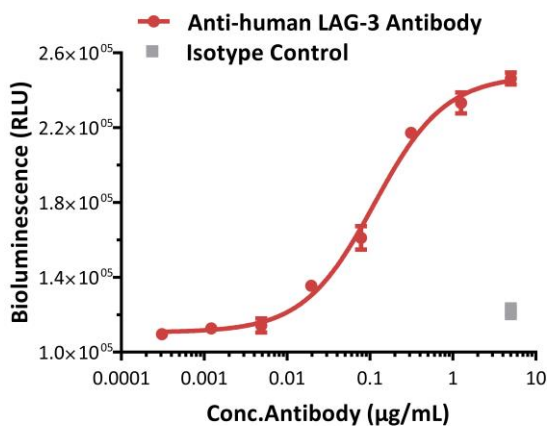
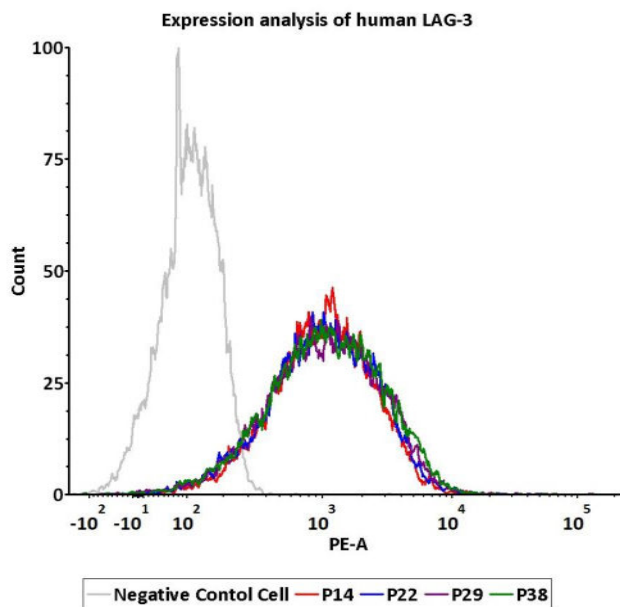


Fig2. Blocking activity of anti-human LAG-3 antibody. This reporter cell was incubated with serial dilutions of antibodies in the presence of target cells expressing human MHCII. The EC₅₀ of anti-human LAG-3 antibody was approximately 0.11 µg/mL.

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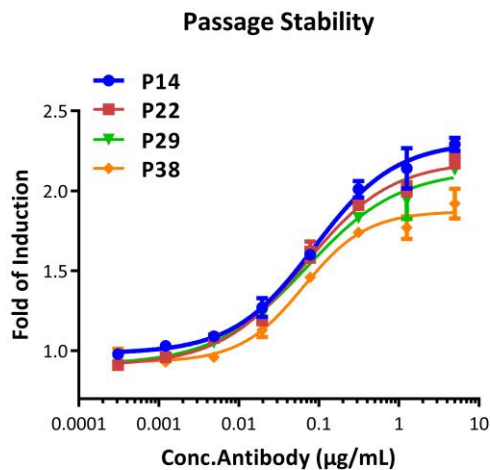
• *Passage Stability*



Passage	MFI for LAG-3 (PE)
P14	992
P22	988
P29	1000
P38	1031

Fig3. Passage stability analysis of receptors expression by FACS. Flow cytometry surface staining of human LAG-3 on Human LAG-3 (Luc) Jurkat Reporter Cell demonstrates consistent mean fluorescent intensity across across passage 14-38.

Human LAG-3 (Luc) Jurkat Reporter Cell Development Service Data Sheet



	P14	P22	P29	P38
EC50 (µg/mL)	0.08706	0.06816	0.0701	0.06419
Max Fold	2.29	2.19	2.13	1.92

Fig4. Passage stability analysis by anti-human LAG-3 antibody stimulation. The continuously growing Human LAG-3 (Luc) Jurkat Reporter Cell was stimulated with serial dilutions of antibodies in the presence of target cells expressing human MHCII. Anti-human LAG-3 antibody stimulated response demonstrates passage stabilization (fold induction and EC50) across passage 14-38.

• License Disclosure

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• Related Products

Products

Human PD-1 (Luc) Jurkat Reporter Cell
 Human TIGIT (Luc) Jurkat Reporter Cell

Cat.No.

SCJUR-STF064
 SCJUR-STF066