Raji/Human CD19 Knockout Stable Cell Line Data Sheet Development Service

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Catalog No.	Size
SCRAJ-STT216	2 × (1 vial contains ~5×10^6 cells)

• *Description*

The Raji/Human CD19 Knockout Stable Cell Line was generated from Raji cells by CRISPR/Cas9-mediated knockout of human CD19 (Gene ID:930). The expression level of human CD19 was confirmed by flow cytometry. Mutated sequences of human CD19 produced by non-homologous end joining (NHEJ) were confirmed through genomic sequencing.

• *Application*

- Useful for cell-based CD19 target-specific analysis

• *Cell Line Profile*

Cell line	Raji/Human CD19 Knockout Stable Cell Line
Host Cell	Raji
Property	suspension
Complete Growth Medium	RPMI Medium 1640 + 10% FBS
Selection Marker	NA
Incubation	37°C with 5% CO_2
Doubling Time	16-20 hours
Transduction Technique	Lentivirus

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• *Materials Required for Cell Culture*

- PRMI-1640 Medium (ATCC, Cat. No. 30-2001™)
- Fetal bovine serum (Gibco, Cat. No. 10091-148)
- Penicillin-Streptomycin (Gibco, Cat. No. 15140-122)
- Phosphate Buffered Saline (1X) (HyClone, Cat. No. SH30256.01)
- Culture Medium: RPMI Medium 1640 + 10% FBS , 1%P/S
- Freeze Medium: 90% FBS, 10% (V/V) DMSO
- T-75 Culture flask (Corning, Cat. No. 430641)
- Cryogenic storage vials (SARSTEDT, Cat. No. 72.379.007)
- Thermostat water bath
- Centrifuge (Cence, Model: L550)
- Cell counter (MONWEI, Model: SmartCell200A Plus)
- CO_2 Incubator (Thermo, Model: 3111)
- Biological Safety Cabinet (Thermo, Model: 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 2 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 culture medium and spin at approximately 1000 rpm for 5 minutes.
4. Resuspend cell pellet with 5 mL culture medium and transfer the cell suspension into T-75 flask containing 10-15 mL of pre-warmed culture medium.
5. Incubate at 37°C with 5% CO_2 incubator until the cells are ready to be split.

• *Subculture*

Adjust the cell density at 1×10^5-2×10^5 viable cells/mL by the addition of fresh medium or replacement of culture medium. Do not allow the cell density to exceed 2×10^6 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

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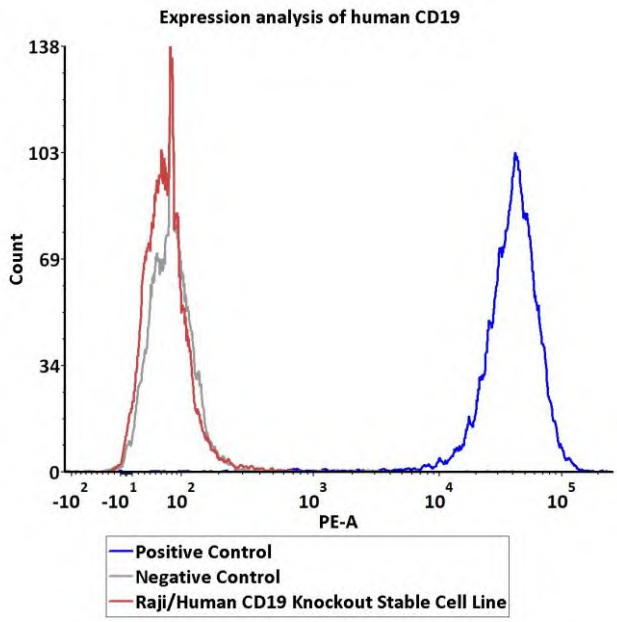
- ***Receptor Assay***



Catalog No.	Stable Cell Line	MFI for CD19 (PE)
NA	Negative Control Cell	74.34
NA	Positive Control Cell	39262.23
SCRAJ-STT216	Raji/Human CD19 Knockout Stable Cell Line	63.92

Fig1. Expression analysis of human CD19 on Raji/Human CD19 Knockout Stable Cell Line by FACS.

Cell surface staining was performed on Raji/Human CD19 Knockout Stable Cell Line using PE-labeled anti-human CD19 antibody. The Raji cells were stained with PE-labeled anti-human CD19 antibody as the positive control cell. The Raji cells were stained with PE-labeled isotype control antibody as the negative control cell.

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- ***Sequencing Analysis***

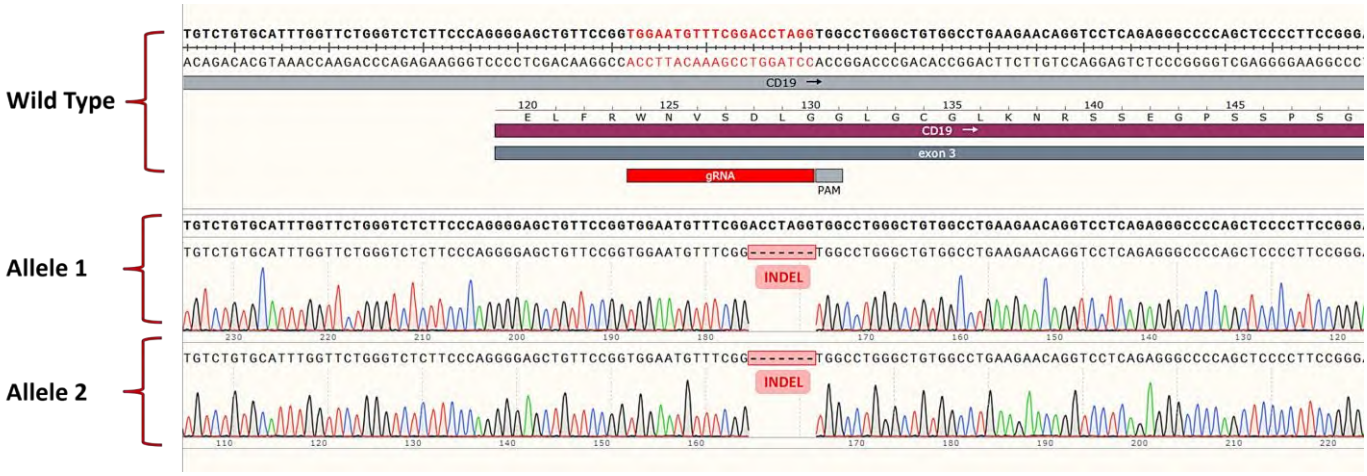

Fig2. Genomic Sequencing of human CD19 in the Raji/Human CD19 Knockout Stable Cell Line.

Sanger sequencing was used for analysis of CRISPR-mediated mutations. The sequencing results demonstrated that the selected sgRNA worked effectively with Cas9 on human CD19 gene in the Raji/Human CD19 Knockout Stable Cell Line.

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• *Related Products*

Products	Cat.No.
Raji/Human PD-L1 Stable Cell Line Development Service	SCRAJ-STT075
Raji/Human CD155 Stable Cell Line Development Service	SCRAJ-STT076