Data Sheet

LUVA Human Mast Cell Line

SCC205

Pack Size: ≥ 1x10^6 viable cells/vial

Store in liquid nitrogen.

FOR RESEARCH USE ONLY

Not for use in diagnostic procedures. Not for human or animal consumption.

Background

Mast cells are a type of granulocyte cell that are important components for the immune system and the process of inflammation. When activated, they will release compounds for inflammation. They are also directly involved in causing allergies and related disorders. Previously, mast cell lines such as LAD2 were used as in vitro options for researching mast cells, however, they were unstable and inconsistent for culturing.[1] A new mast cell line was developed and given the name of LUVA cells, which mirror the morphology and functionality of human mast cells.

The CD34+ mast cells were cultured for 8 weeks with stem cell factor and cultured with IL-6 and IL-3 for only one week. The stable LUVA cells were maintained and became capable of growing without further addition of growth factors in culture, making them highly convenient for culturing. They are the first mast cell line in which the donor patient did not have a mast cell disorder, as well as having no mutation in the c-kit receptor.[1]

The LUVA cell line allows for a proper in vitro mast cell model in which researchers can study the mechanisms which cause mastocytosis, a condition where mast cells aggregate in certain parts of the body.[2] This condition results in a higher risk of life-threatening allergic reactions as well as rarely resulting in mast cell cancers. Commonly, mastocytosis is caused by a mutation of the c-KIT protein, a receptor tyrosine kinase. By understanding c-KIT mechanisms, therapies can continue to develop and improve for mastocytosis patients.

Source

The LUVA mast cell line was derived from CD34+ cells of a non-mastocytosis donor with aspirin-exacerbated respiratory disease.

Short Tandem Repeat

D3S1358: 16, 19	D7S820: 11	vWA: 18	FGA: 23, 24
D8S1179: 13, 14	D21S11: 32.2, 33.2, 34.2	D18S51: 13, 14	D5S818: 10, 11
D13S317: 10, 12	D16S539: 11	TH01: 7	TPOX: 11
CSF1PO: 9, 10	AMEL: X, Y	Penta D: 13, 14	Penta E: 5, 10

Quality Control Testing

- LUVA Human Mast Cells are verified to be of human origin and negative for mouse, rat, Chinese hamster, Golden Syrian hamster, and nonhuman primate interspecies contamination, as assessed by a Contamination Clear panel by Charles River Animal Diagnostic Services.
- Cells tested negative for infectious diseases against a Human Essential CLEAR panel by Charles River Animal Diagnostic Services.
- Cells tested negative for mycoplasma.

Storage and Handling

LUVA cells should be stored in liquid nitrogen until use. The cells can be cultured for at least 10 passages after initial thawing without significantly affecting the cell marker expression and functionality.

Representative Data

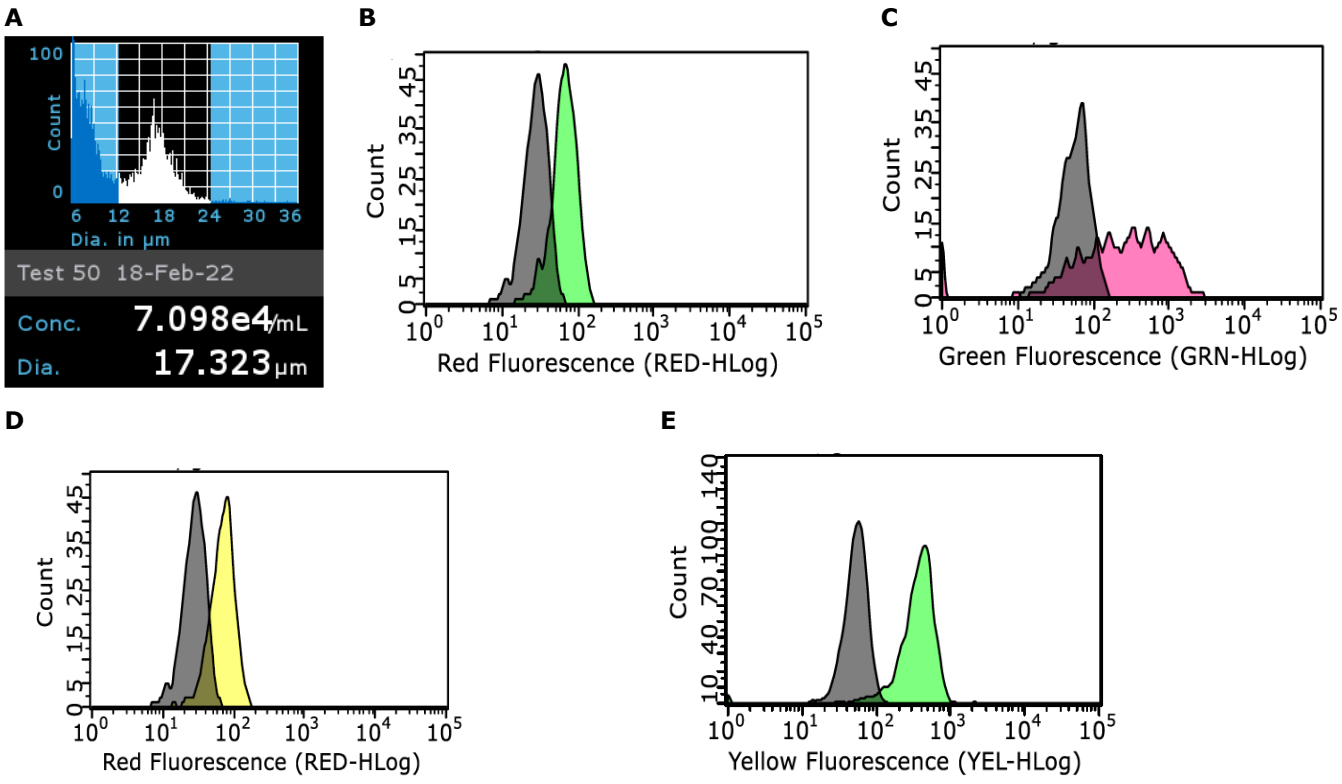

Figure 1. (**A**) Cell counting was performed using the Scepter™ 3.0 Handheld Automated Cell Counter using 60µm sensors (PHCC360KIT). (**B**) LUVA cells were characterized by flow cytometry and shown to express TPSAB1 (SAB1303861), (**C**) C-Kit (SAB4700711), (**D**) CD32 (ZRB1275), and (**E**) FceR1 (Invitrogen,12-5899-42).

Protocols

Thawing the Cells

1. Do not thaw the cells until the recommended medium is on hand. Cells can grow on standard tissue cultureware surfaces without any additional coating.

   LUVA cells are thawed and expanded in LUVA Expansion Medium comprising of StemPro®-34 SFM Medium+Nutrient Supplement (ThermoFisher, 10639011) containing 2 mM L-Glutamine (G7513) and Penicillin/Streptomycin (P4333) (optional).
2. Remove the vial of frozen LUVA cells from liquid nitrogen and incubate in a 37 °C water bath. Closely monitor until the cells are completely thawed. Maximum cell viability is dependent on the rapid and complete thawing of frozen cells.

   IMPORTANT: Do not vortex the cells.
3. As soon as the cells are completely thawed, disinfect the outside of the vial with 70% ethanol. Proceed immediately to the next step.
4. In a laminar flow hood, use a 1-2 mL pipette to transfer the cells to a sterile 15 mL conical tube. Be careful not to introduce any bubbles during the transfer process.
5. Using a 10 mL pipette, slowly add dropwise 9 mL of LUVA Expansion Medium (Step 1 above) to the 15 mL conical tube.

   IMPORTANT: Do not add the entire volume of media all at once to the cells. This may result in decreased cell viability due to osmotic shock.
6. Gently mix the cell suspension by slowly pipetting up and down twice. Be careful not to introduce any bubbles.

   IMPORTANT: Do not vortex the cells.
7. Centrifuge the tube at 300 x *g* for 2-3 minutes to pellet the cells.
8. Decant as much of the supernatant as possible. Steps 5-8 are necessary to remove residual cryopreservative (DMSO).
9. Resuspend the cells in 6 mL of LUVA Expansion Medium.
10. Transfer the cell mixture to a T25 tissue culture flask. Incubate the cells at 37 °C in a humidified incubator with 5% CO_2.

    Note: LUVA cells should be cultured with the flask standing upright. Undesired cell adherence may result if cells are grown with flask laid flat.

Subculturing the Cells

1. LUVA cells are a suspension cell line. They should be passaged before reaching 1,000,000 cells/mL. The cells are recommended to be subcultured to a minimum cell density of 300,000 cells/mL. LUVA cells require media changes every 2-3 days.
2. To change media, centrifuge the tube at 300 x *g* for 3-5 minutes to pellet the cells.
3. Discard the supernatant, then loosen the cell pellet by tapping the tip of the tube with a finger.
4. Add desired volume of LUVA Expansion Medium to the conical tube and resuspend the cells thoroughly. Large cell clumps may be broken up by gentle trituration.

   IMPORTANT: Do not vortex the cells.
5. Count the number of cells using a hemocytometer or a Scepter™ 3.0 Handheld Automated Cell Counter.
6. Plate the cells to the desired density.

Cryopreservation of the Cells

LUVA cells may be frozen in CryoStor®10 cell cryopreservation media (C2874).

References

1. Laidlaw TM, Steinke JW, Tiñana AM, Feng C, Xing W, Lam BK, Paruchuri S, Boyce JA, Borish L. 2011. Characterization of a novel human mast cell line that responds to stem cell factor and expresses functional FcεRI. J Allergy Clin Immunol. 127(3): 815-822.
2. Arock M, Wedeh G, Hoermann G, Bibi S, Akin C, Peter B, Gleixner KV, Hartmann K, Butterfield JH, Metcalfe DD, et al. 2018. Preclinical human models and emerging therapeutics for advanced systemic mastocytosis. Haematologica. 103(11): 1760-1771.

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