



# SNCA3x\_2KO\_C2

SNCA3x\_2KO\_C2<sup>TM</sup>

## Description

SNCA3x\_0KO\_C1 is a CRISPR-engineered induced pluripotent stem cell (iPSC) with SNCA frameshift mutations from a donor with an SNCA triplication. This cell line is part of a panel of cells that can be used to study alpha-synuclein gene expression dosage from the endogenous locus and has applications in Parkinson's disease research.

**Organism:** *Homo sapiens*, human

**Tissue:** Skin

**Age:** 42 years

**Gender:** Male

**Growth properties:** Adherent

**Disease:** Parkinsons disease

**Cells per vial:**  $\geq 1.0 \times 10^6$

**Volume:** 1.0 mL

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## Storage Conditions

**Product format:** Frozen

**Storage conditions:** Vapor phase of liquid nitrogen

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## Intended Use

This product is intended for laboratory research use only. It is not intended for any animal or human therapeutic use, any human or animal consumption, or any diagnostic use.

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## BSL 2



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ATCC determines the biosafety level of a material based on our risk assessment as guided by the current edition of *Biosafety in Microbiological and Biomedical Laboratories (BMBL)*, U.S. Department of Health and Human Services. It is your responsibility to understand the hazards associated with the material per your organization's policies and procedures as well as any other applicable regulations as enforced by your local or national agencies.

ATCC highly recommends that appropriate personal protective equipment is always used when handling vials. For cultures that require storage in liquid nitrogen, it is important to note that some vials may leak when submersed in liquid nitrogen and will slowly fill with liquid nitrogen. Upon thawing, the conversion of the liquid nitrogen back to its gas phase may result in the vial exploding or blowing off its cap with dangerous force creating flying debris. Unless necessary, ATCC recommends that these cultures be stored in the vapor phase of liquid nitrogen rather than submersed in liquid nitrogen.

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### Certificate of Analysis

For batch-specific test results, refer to the applicable certificate of analysis that can be found at [www.atcc.org](http://www.atcc.org).

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### Growth Conditions

**Temperature:** 37°C

**Atmosphere:** 95% Air, 5% CO<sub>2</sub>

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### Handling Procedures

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#### Complete medium:

The basal medium for this cell line is StemFlex™ Basal Medium. To make the complete medium add the following to 450 mL basal medium:

- 50 mL StemFlex™ Supplement 10X
- 1  $\mu$ M of Thiazovivin: 1 mM Thiazovivin/DMSO solution - prepared using 1 mg Thiazovivin/ 3.1212 ml DMSO

**Note:** Thiazovivin/DMSO solution is used only at Start-Up and Day 0 of all subcultures and removed next day - substituted with StemFlex™ Basal Medium.

#### Subculturing procedure:

Volumes used in this protocol are for 75 cm<sup>2</sup> flask; proportionally reduce or increase amount of dissociation medium for culture vessels of other sizes. Corning® T-75 flasks (catalog #430641) are recommended for subculturing this product.

Notes: CBM coated culture vessels used. Do NOT use CellSTACKs Note on dissociation medium

1. Remove and discard culture medium.
2. Briefly rinse the cell layer with PBS.
3. Add 2.0 to 3.0 mL of 0.5mM EDTA solution to flask and observe cells under an inverted microscope until cell layer is dispersed (usually within 5 to 15 minutes).

Note: To avoid clumping do not agitate the cells by hitting or shaking the flask while waiting for the cells to detach. Cells that are difficult to detach may be placed at 37°C to facilitate dispersal.

4. Add 6.0 to 8.0 mL of complete growth medium and aspirate cells by gently pipetting.

5. **Centrifuge cells to remove dissociation agent and resuspend in complete media .**

6. Add appropriate aliquots of the cell suspension to new CBM coated culture vessels.

Cultures can be established between  $2 \times 10^4$  and  $1 \times 10^5$  viable cells/cm<sup>2</sup>. Do not exceed  $7 \times 10^4$  cells/cm<sup>2</sup>.

7. Incubate cultures at 37°C.

**Interval:** Maintain cultures at a cell concentration between  $1.5 \times 10^4$  and  $6 \times 10^4$  cell/cm<sup>2</sup>.

**Subcultivation Ratio:** A subcultivation ratio of 1:3 to 1:8 is recommended

**Medium Renewal:** 2 to 3 times per week

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**Notes:** Prepare in advance CBM coated cell culture flasks prior to subculturing. Media changes must be performed every other day. Subculture at  $\leq$  70% confluence. For all startups dilute 1 mL of cell vial with 4 mL of complete medium and perform cell counts prior to centrifugation.

**Reagents for cryopreservation:** BAMBANKER™ Serum Free Cell Freezing Medium (Fisher Scientific catalog # NC9582225)

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## Material Citation

If use of this material results in a scientific publication, please cite the material in the following manner: SNCA3x\_2KO\_C2 (ATCC SNCA3x\_2KO\_C2)

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## References

References and other information relating to this material are available at [www.atcc.org](http://www.atcc.org).

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#### **Revision**

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