Data Sheet

Oli-neu Mouse Oligodendrocyte Precursor Cell Line

Immortalized Cell Line

SCC261

Pack Size ≥1x10^6 viable cells/vial

Store at: Liquid Nitrogen

FOR RESEARCH USE ONLY

Not for use in diagnostic procedures. Not for Human or Animal Consumption.

Background

Oligodendrocytes, comprising 5-10% of the total population of glial cells in the central nervous system, are responsible for producing the myelin sheath encasing axons which is essential for efficient electric conduction of neural impulses.[1] Oligodendrocytes arise from precursor cells that differentiate just before birth. Disorders of myelination of axons can lead to emergence of neurodegenerative diseases such as Alzheimer's and multiple sclerosis, underscoring the critical importance of understanding oligodendrocyte biology and interactions. The myelination process occurs within the complex milieu of neuronal cell types, thus is difficult to reconstruct without cellular models.

Oli-neu is an immortalized mouse oligodendroglial cell line that recapitulates morphology and *in vivo* physiological activity of oligodendroglial cells. Oli-neu cells differentiate into oligodendrocytes upon daily addition of 1 mM dibutyrl-cAMP.[2] Oli-neu cells have been utilized in a broad range of studies of oligodendrocyte biology and has greatly contributed to the understanding of the mechanisms of myelination and molecular players in myelination disorders. Differentiated Oli-neu oligodendrocytes express O4 antigen, an oligodendrocyte marker, and manifest cell processes characteristic of oligodendrocyte morphology.[2] The Oli-neu cell line is a versatile cellular model for oligodendrocyte biology and associated neurophysiological investigations.

Source

Oli-neu was derived from oligodendrocytes and oligodendrocyte precursor cells isolated from the cerebellum of a 6-day-old NMRI mouse and infected with retrovirus containing the t-neu tyrosine kinase oncogene.[2]

Short Tandem Repeat

M18-3: 17	M15-3: 20.3
M4-2: 19.3	M6-4: 16, 17, 18
M6-7: 11, 12	M11-2:15
M19-2: 11, 13	M17-2:15, 16
M1-2: 14	M12-1:19, 20
M7-1: 25.2, 29	M5-5: 12
M1-1: 14, 15	MX-1: 25
M3-2: 10, 13	M13-1: 15
M8-1: 16	D8S1106: NA
M2-1: 9	D4S2408: NA

Quality Control Testing

- Each vial contains ≥ $1x10^6$ viable cells.
- Cells are tested negative for infectious diseases by a Mouse Essential CLEAR Panel by Charles River Animal Diagnostic Services.
- Cells are verified to be of mouse origin and negative for inter-species contamination from rat, human, Chinese hamster, Golden Syrian hamster, and Non-human Primate (NHP) as assessed by a Contamination Clear panel by Charles River Animal Diagnostic Services.
- Cells are negative for mycoplasma contamination.

Storage and Handling

Oli-neu mouse oligodendrocyte precursor cells should be stored in liquid nitrogen. The cells can be cultured for at least 10 passages after initial thawing without significantly affecting functionality.

Representative Data

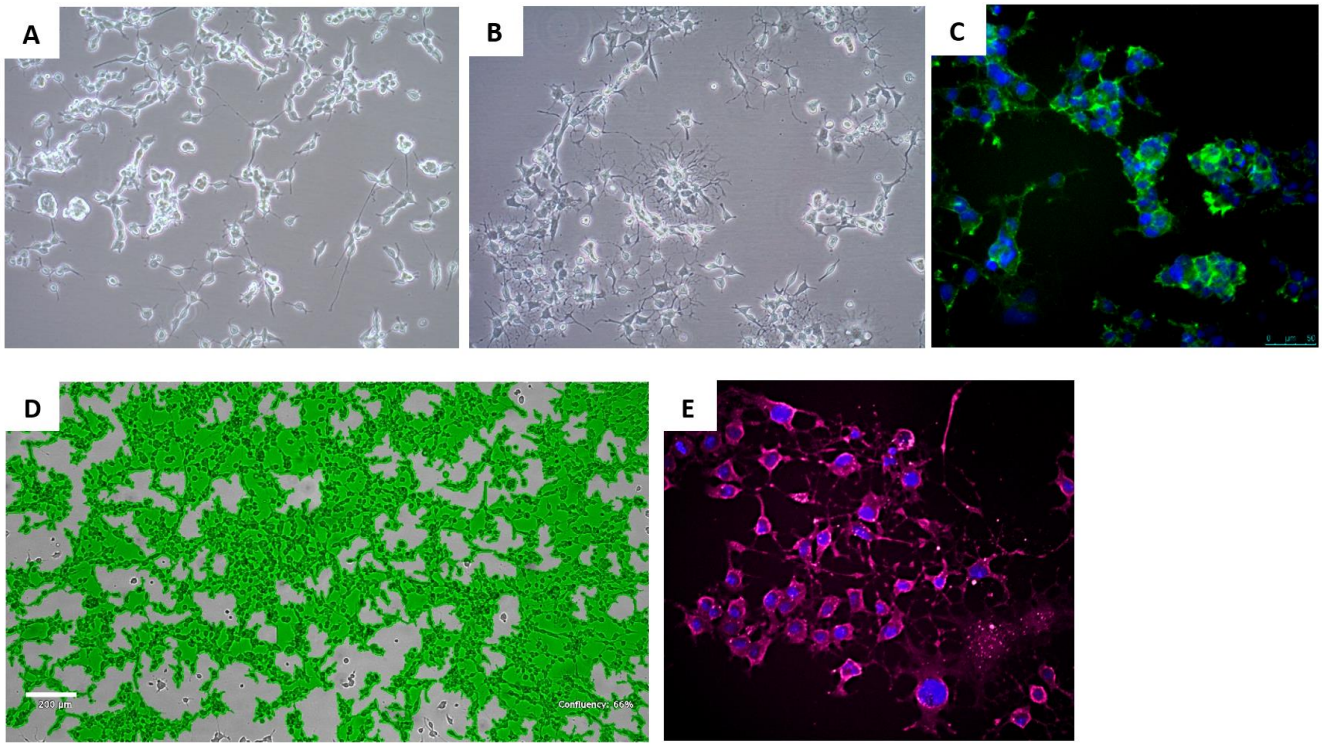

Figure 1. Brightfield images of oli-neu cells undifferentiated (**A**) and differentiated (**B**) for 4 days with 1 mM dibutyrl cAMP. Actin staining (green) of undifferentiated cells (**C**). Cell confluency (66%) was assessed using MilliCell® Digital Cell Imager (**D**, Cat. No. MDCI 10000). Differentiated cells express O4 oligodendrocyte marker (**E**, red, Cat. No. 07139).

Protocols

Thawing Cells

1. **Prepare poly-L-lysine coated flasks:** Coat flasks overnight at room temperature with poly-l-lysine 0.01% solution (Cat. No. A-005-M). When cells are ready to be plated, aspirate the coating solution and rinse twice with sterile water before plating Oli-neu cells.
2. **Prepare Oli-neu Expansion Medium:** Comprised of DMEM High Glucose Medium (Cat. No. D5796) containing 10 µg/mL Insulin (Cat. No. I9278-5ML), 10 µg/mL Apo-transferrin (Cat. No. T4382), 1X B27™ Supplement (50X) (ThermoFisher 17504044), 100 µM Putrescine dihydrochloride (Cat. No. P5780), 200 nM Progesterone (Cat. No. P0130), 500 nM 3,3’,5-Triiodo-L-thyronine (Cat. No. T6397), 520 nM L-Thyroxin (Cat. No. T1775), and 220 nM Sodium Selenite (Cat. No. S5261).

   Note: Expansion medium should not contain FCS as this may push the cells towards the astroglial lineage.
3. Remove the vial of frozen Oli-neu 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.
4. As soon as the cells are completely thawed, disinfect the outside of the vial with 70% ethanol. Proceed immediately to the next step.
5. In a laminar flow hood, use a 1- or 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.
6. Using a 10 mL pipette, slowly add dropwise 9 mL of cold Oli-neu Expansion Medium (Step 2 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.

7. 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.
8. Centrifuge the tube at 2-8 °C at 300 *x g* for 2-3 minutes to pellet the cells. Decant as much of the supernatant as possible. Steps 5-8 are necessary to remove residual cryopreservative (DMSO).
9. Resuspend the cells in 10-15 mL of Oli-neu Expansion Medium. Transfer the cell mixture to a poly-l-lysine coated T75 tissue culture flask. Incubate the cells at 37 °C in a humidified incubator with 5% CO_2. Exchange medium every other day.
10. Oli-neu should be passaged at ~80-85% confluency to a new poly-l-lysine coated flask. Typical split ratio is 1:6. Oli-neu cells usually do not grow as dense as for example HEK cells. Thus, at full confluency, there will be some space between the cells. Monitor the cells carefully as they may change growing behavior from time to time. Adjust the plating density accordingly (i.e. slowly growing cells have to be seeded at a higher density and vice versa).

Oli-neu Differentiation

1. Differentiation can be induced by daily addition of 1 mM dibutyrl cAMP. Maximal differentiation is estimated after one week of treatment, but normally 3-4 days are sufficient to achieve considerable differentiation. A stop in treatment or medium change may lead to de-differentiation.
2. Prepare a subconfluent 10-cm plate by plating proximately 4-5 million cells per plate.
3. Initiate differentiation by adding dibutyrl cAMP to 1 mM final concentration.

Cryopreservation of Cells

Oli-neu cells may be frozen in RPMI-1640 medium containing 20% FBS and 10% DMSO using a Nalgene® slow freeze Mr. Frosty® container.

References

1. Curr Pharm Des. 2016, 22(6): 656-679.
2. Eur J Neurosci. 1995, 7(6): 1245-1265.

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