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User Manual

OriCell™ Sprague-Dawley (SD) Fetal Rat Hippocampus Neurons

Cat. No. SHCFN-00001

Table of Contents

CONTENTS AND STORAGE

Product Name	Sprague-Dawley Fetal Rat Hippocampus Neurons
Catalog No.	SHCFN-00001
Amount per Vial	1×10^6 Cells
Cryopreserved At	Primary Passage
Storage Condition	Liquid Nitrogen

CAUTION: **Please handle this product as a potentially biohazardous material. This product contains Dimethyl Sulfoxide (DMSO), a hazardous material, in the freezing medium.**

PRODUCT INTRODUCTION

Neurons, also known as nerve cells, connect to each other to form networks. They are the main components of the nervous system and are potent tools in studying function of the neural system.

Cyagen OriCell™ Sprague-Dawley (SD) Fetal Rat Hippocampus Neurons are derived from the Hippocampus of qualified SD rat embryos (18.5 dpc) and cryopreserved as primary cells. These cells express specific clusters of different proteins for neurons. They have been tested negative for bacteria, fungi and mycoplasma.

Viability of Cyagen OriCell™ SD Fatal Rat Hippocampus Neurons

- Recovery Viability ≥ 50%

Purity of Cyagen OriCell™ SD Fatal Rat Hippocampus Neurons

- MAP2 positive cells ≥ 70%
- β-tubulin III positive cells ≥ 70%
- GFAP positive cells ≤ 10%

This product is intended for laboratory research use only. It is not intended for diagnostic, therapeutic, clinical, household, or any other applications.

PRODUCT APPLICATIONS

OriCell™ Sprague-Dawley (SD) Fetal Rat Hippocampus Neurons can be used in neurobiology studies and drug discovery in areas such as Parkinson's Disease and Alzheimer's Disease.

GENERAL HANDLING PRINCIPLES

Aseptic handling of the product is necessary throughout.

POLY-L-LYSIN/LAMININ COATING OF TISSUE CULTURE PLATE

1. The day before plating cells for differentiation, prior to prepare the coated plates with PLL/laminin.
2. Dilute Poly-L-lysine stock Solution (1mg/mL) with water to yield 15µg/mL solution.
3. Add enough of Poly-L-lysine Solution into the culture vessel to completely cover the base.
4. Swirl until Poly-L-lysine Solution coats the entire base of vessel. Sit for at least 30 minutes at room temperature.
5. Aspirate off all of the Poly-L-lysine Solution and rinse the vessel once with sterile water. Aspirate after each rinse.
6. Use sterile 1X PBS, dilute the Laminin stock Solution (1mg/mL) to a final concentration of 15µg/mL.
7. Add enough of Laminin Solution into the culture vessel to completely cover its base. Incubate overnight at 4°C, coated vessels can be stored in the Laminin Solution at 4°C for up to one week.
8. Just before use, aspirate the Laminin Solution in the coated vessels and wash the wells once with 1X PBS. Aspirate after rinse.

THAWING AND ESTABLISHING OriCell™ SPRAGUE-DAWLEY(SD) FETAL RAT HIPPOCAMPUS NEURONS

Materials Required:

- OriCell™ Sprague-Dawley Fetal Rat Hippocampus Neurons (Cat. No. SHCFN-01001)
- OriCell™ Neuron Growth Medium (Cat. No. GXXNR-90011)

Thawing and Establishing SD Fetal Rat Hippocampus Neurons

1. Pre-warm the OriCell™ SD Fatal Rat Hippocampus Neuron Growth Medium to 37°C.
2. Remove the cryovial of OriCell™ SD Fatal Rat Hippocampus Neurons from liquid nitrogen.

3. Quickly thaw the vial in a 37°C water bath until the last ice crystal disappears. For optimal results, be sure to finish the thawing procedure within 3 minutes. Be careful not to submerge the entire vial. Maximum cell viability is dependent on the rapid and complete thawing of frozen cells.

Note: **Thawing the cells for longer than 3 minutes results in less than optimal results.**

4. As soon as the cells are completely thawed, disinfect the outside of the vial with 70% v/v ethanol.
5. Use a pipette to transfer 1mL Neurons Growth Medium to the vial drop by drop in side a biosafety cabinet, mix by gently pipetting. Subsequently transfer this 2mL of cell suspension into the conical tube containing Neurons Growth Medium drop by drop. Be careful not to introduce any bubbles during the transfer process.
6. Rinse the vial with 1 mL of medium to reduce cell loss. Subsequently transfer this 1mL of cell suspension to the conical tube.
7. Gently mix the cell suspension by slowly pipetting up and down. Be careful not to introduce any bubbles.
8. Take a small sample of the cells from the cells suspension. Determine the viable cell density using vital staining assay with trypan blue.
9. Plate around 1×10^5 /cm^2 viable cells in poly-L-lysine/Laminin coated plate. Gently rock the culture plate to evenly distribute the cells.

Note: **Do not centrifuge the cells as they are extremely fragile upon recovery.**

10. Incubate the cells at 37°C in a 5% CO_2 humidified incubator.
11. Six hours later, change half of the medium with fresh OriCell™ Neuron Growth Medium (pre-warmed to 37°C), Change half of the medium every three days, or change the entire medium every three days if there is any cell death.

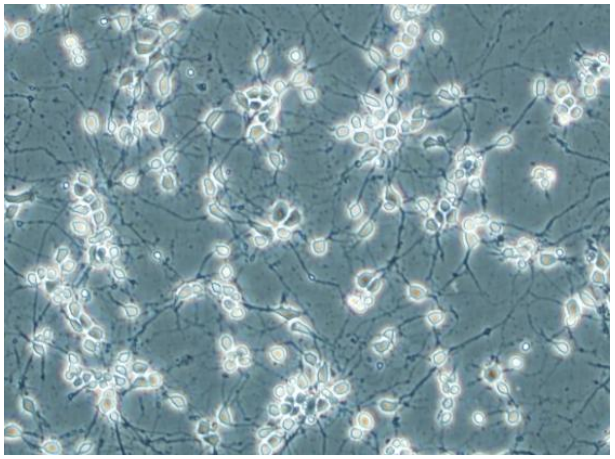

Fig. 1 OriCell™ SD Fetal Rat Hippocampus Neurons are established

APPENDIX

Related products

Product	Catalog Number
OriCell™ Sprague-Dawley Fetal Rat Hippocampus Neurons	SHCFN-00001
OriCell™ Sprague-Dawley Fetal Rat Cortex Neurons	SCCFN-00001
OriCell™ Neuron Growth Medium	GXXNR-90011

References

Greory J Brewer, and John R Torricelli. (2007) Isolation and culture of adult neurons and neurospheres. *Nature* 2: 1490-1498.

CHENGSONG XIE, William R, Markesbery, and Mark A, Lovell. (2000) Survival of Hippocampal and Cortical Neurons in a Mixture of MEM and B27 Supplemented Neurobasal Medium. *Free Radical Biology & Medicine* 28: 665-672.