

**Mouse Neurons-raphe** 

(MN-r)

Catalog Number: M1510-57

# **Cell Specification**

The tissue of the central nervous system is made up of two classes of cells that may be broadly categorized as neurons and glia. Neurons are anatomic, functional, and trophic units of the brain [1]. Despite great variability in size and shape, all neurons share common morphological features, which are those of the key elements of a highly complex communication network. The neurons are the dynamically polarized cells that serve as the major signaling unit of the nervous system.

The MN-r from ScienCell Research Laboratories are isolated from E14 C57BL/6 mouse brain raphe. MN-r are cryopreserved at primary cultures and delivered frozen. Each vial contains >1 x  $10^6$  cells in 1 ml volume. MN-r are characterized by immunofluorescent method with antibodies to neurafilament, MAP2, and beta-tubulin III. MN-r are negative for mycoplasma, bacteria, yeast and fungi. MN-r are guaranteed to further culture in the conditions provided by ScienCell Research Laboratories.

#### **Recommended Medium**

It is recommended to use Neuronal Medium (NM, Cat. No. 1521) for the culturing of MN-r in vitro.

#### **Product Use**

MN-r are for research use only. They are not approved for human or animal use, or for application in *in vitro* diagnostic procedures.

# **Storage**

Transfer cells directly and immediately from dry ice to liquid nitrogen upon receiving and keep the cells in liquid nitrogen until cell culture is needed for experiments.

## Shipping

Dry ice.

#### Reference

- [1] Parent, A. (1996) Neurons in Carpenter's Human Neuroanatomy. 9th ed., pp131-198, Williams & Wilkins, Quebec, Canada.
- [2] Alberts, B., Bray, D., Lewis, J., Raff, M., Roberts, M., Watson, J. D. (1989) Molecular biology of the cell. 2nd. ed., New York: Garland.

# **Instruction for culturing cells**

Caution: Cryopreserved cells are very delicate. Thaw the vial in a 37°C waterbath

and return them to culture as quickly as possible with minimal handling!

# Set up culture after receiving the order:

1. Coat culture vessel with 0.2% Gelatin (ScienCell Product # 0423)

Note: It is important that these cells are plated in Gelatin coated culture vessels that promote cell attachment (Gelatin coating: coat flask or plate with 0.2% Gelatin at suggested volumes for one hour and wash the flask or plate with sterile water three times).

- 1. Prepare complete medium: decontaminate the external surfaces of medium and medium supplements with 70% ethanol and transfer them to sterile field. Aseptically open each supplement tube and add them to the basal medium with a pipette. Rinse each tube with medium to recover the entire volume.
- 2. Add 7 ml of complete medium to the flask. Leave the flask in the hood and go to thaw the cells.
- 3. Place the vial in a 37°C waterbath, hold and rotate the vial gently until the contents are completely thawed. Remove the vial from the waterbath immediately, wipe it dry, rinse the vial with 70% ethanol and transfer it to a sterile field. Remove the cap, being careful not to touch the interior threads with fingers. Using 1 ml eppendorf pipette gently resuspend the contents of the vial.
- 4. Dispense the contents of the vial into the equilibrated, gelatin coated culture vessels. A seeding density of  $\geq 10,000$  cells/cm<sup>2</sup> is recommended.
  - Note: Dilution and centrifugation of cells after thawing are not recommended since these actions are more harmful to the cells than the effect of DMSO residue in the culture. It is also important that cells are plated in gelatin coated culture vessels that promote cell attachment.
- 5. Replace the cap or cover, and gently rock the vessel to distribute the cells evenly. Loosen cap if necessary to permit gas exchange.
- 6. Return the culture vessels to the incubator.
- 7. For best result, do not disturb the culture for at least 16 hours after the culture has been initiated. Change the growth medium the next day to remove the residual DMSO and unattached cells, then every other day thereafter.

### **Maintenance of Culture:**

- 1. Change the medium to fresh supplemented medium the next morning after establishing a culture from cryopreserved cells.
- 2. Change the medium every two to three days thereafter.

# It is not recommended that MN-r be subcultured beyond their initial plating.

Caution: Handling animal derived products is potentially biohazardous. Always wear gloves and safety glasses when working these materials. Never mouth pipette. We recommend following the universal procedures for handling products of animal origin as the minimum precaution against contamination [1].

[1]. Grizzle, W. E., and Polt, S. S. (1988) Guidelines to avoid personal contamination by infective agents in research laboratories that use human tissues. J Tissue Culture Methods. 11(4).