Human Neurons-brain stem (HN-bs)

Catalog Number: 1560

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. Neurons are dynamically polarized cells that serve as the major signaling unit of the nervous system. The brain is known to contain about 1×10^{11} neurons, each being able to contact at least 10,000 other neurons [2]. Brain stem neurons provide main motor and sensory innervation and integrative functions by regulating heart rate, breathing, sleep cycle, awareness, consciousness, and more [3-4]. Neurons isolated from different brain regions serve as excellent models for studies of region-specific pathologies and degeneration, neurotoxicology and brain development [5-7].

HN-bs from ScienCell Research Laboratories are isolated from human brain stem tissue. HN-bs are cryopreserved at primary culture and delivered frozen. Each vial contains $>1 \times 10^6$ cells in 1 ml volume. HN-bs are characterized by immunofluorescence with antibodies specific to neurofilament, MAP2, and beta-tubulin III. HN-bs are negative for HIV-1, HBV, HCV, mycoplasma, bacteria, yeast and fungi. HN-bs 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 HN-bs i*n vitro*.

Product Use

HN-bs 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., *et. al*. (1989) Molecular biology of the cell. 2nd ed., New York: Garland.

[3] Smith, J., Smith, A, Abdala, H., Koizumi, I., *et al.* (2008) J Neurophysiol. 98: 3370–87.

[4] Zheng, Z., Lewis, M., Alberto Travagli, R. (2005) Am J Physiol Gastrointest Liver Physiol. 288: G1066–73.

[5] Mitchell, J., Paul, P., Chen, H., Morris, A., *et. al*. (2010) Proc Natl Acad Sci U S A. 107:7556–61.

[6] Wang, C., Sadovova, N., Ali, H.K., Duhart, H.M., *et. al*. (2007) Neuroscience. 144:46-55.

[7] Sato, H., Fukutani, Y., Yamamoto, Y., Tatara, E., *et. al*. (2012) J Neurosci. 32:15388-402.

Instruction for culturing cells

Caution: Cryopreserved cells are very delicate. Thaw the vial in a 37°C water bath and return the cells to culture as quickly as possible with minimal handling!

Initiating the culture:

1. Coat culture vessel with 0.2% gelatin (Cat. No. 0423). Recommended to use a T-25 flask (alternatively 3 wells of a 6-well plate or 12 wells of a 24-well plate can be used.) Use enough volume of gelatin to cover the entire culture surface. Wash the vessel with sterile water for three times prior use.

   Note: It is important that these cells are plated in gelatin coated culture vessels to promote cell attachment.

2. Prepare complete medium. Decontaminate the external surfaces of medium bottle and medium supplement tubes with 70% ethanol and transfer them to a sterile field. Aseptically transfer supplement to the basal medium with a pipette. Rinse the tube with medium to recover the entire volume.

3. Add complete medium to the culture vessel. Leave the vessel in the sterile field and proceed to thaw the cryopreserved cells.

4. Place the frozen vial in a 37°C water bath. Hold and rotate the vial gently until the contents completely thaw. Remove the vial from the water bath promptly, wipe it down with 70% ethanol and transfer it to the sterile field. Remove the cap carefully without touching the interior threads.

5. Gently resuspend and dispense the contents of the vial into the equilibrated, gelatin coated culture vessel. A seeding density of ≥100,000 cells/cm^2 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 residual DMSO in the culture. It is also important that cells are plated in gelatin coated culture vessels to promote cell attachment.

6. Replace the cap or lid, and gently rock the vessel to distribute the cells evenly. Loosen cap if necessary to allow gas exchange.

7. Return the culture vessel to the incubator.

8. For the best result, do not disturb the culture for at least 16 hours after the culture has been initiated. Refresh culture medium the next day to remove the residual DMSO and unattached cells, then every other day thereafter.

Maintaining the culture:

1. Refresh supplemented culture 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 neurons be subcultured beyond their initial plating.

Caution: Handling human derived products is potentially biohazardous. Although each cell strain tests negative for HIV, HBV and HCV DNA, diagnostic tests are not necessarily 100% accurate, therefore, proper precautions must be taken to avoid inadvertent exposure. Always wear gloves and safety glasses when working these materials. Never mouth pipette. We recommend following the universal procedures for handling products of human origin as the minimum precaution against contamination [1].

[1] Grizzle WE, Polt S. (1988) "Guidelines to avoid personal contamination by infective agents in research laboratories that use human tissues." *J Tissue Cult Methods.* 11: 191-9.