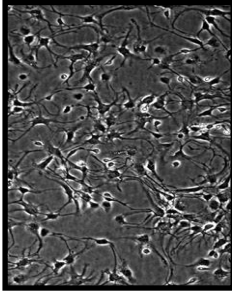


NEUROSCIENCES INNOPROFILE™ HUMAN MESENCEPHALON NEURONS



Product Type:	Cryo-preserved Neurons
Catalog Number:	P10154
Source:	Human Mesencephalon (midbrain)
Number of Cells:	1 x 10 ⁶ Neurons / 1ml
Storage:	Liquid Nitrogen

Human Mesencephalon Neurons from Innoprot are isolated by ScienCell Research Laboratories from human brain midbrain (mesencephalon). Human Mesencephalon Neurons are cryopreserved at Po and delivered frozen. HNMs are guaranteed to further culture in the conditions provided by Innoprot.

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. Despite great variability in size and shape, all neurons share common morphologic 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 signalling unit of the nervous system.

Recommended Medium

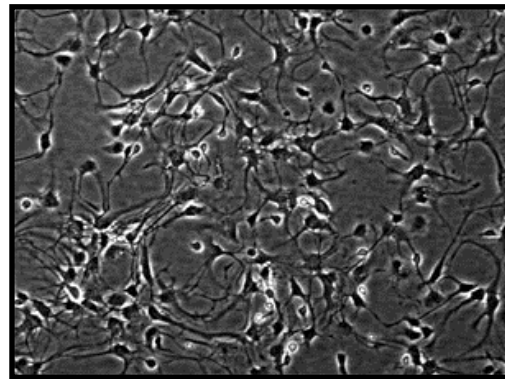
- Neuronal Medium (*serum-free*)
(Reference: P60157)

Product Characterization

Immunofluorescent method

- Neurofilament
- MAP-2
- beta-tubulin III

The cells test negative for HIV-1, HBV, HCV, mycoplasma, bacteria, yeast and fungi



Product Use

THESE PRODUCTS ARE FOR RESEARCH USE ONLY. Not approved for human or veterinary use, for application to humans or animals, or for use in vitro diagnostic or clinical procedures

INSTRUCTIONS FOR CULTURING CELLS

IMPORTANT: 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. Prepare a poly-L-lysine-coated culture vessel (2 µg/cm², T-25 flask is recommended). Add 5 ml of sterile water to a T-25 flask and then add 50 µl of poly-L-lysine stock solution (1 mg/ml, Ref.PLL). Leave the vessel in a 37°C incubator overnight (or for a minimum of one hour). Rinse the poly-L-lysine-coated vessel twice with sterile water prior to use.

It is important that these cells are plated in poly-L-lysine-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 supplement 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. Promptly remove the vial from the water bath, wipe it down with 70% ethanol, and transfer it to the sterile field. Carefully remove the cap without touching the interior threads.

5. Gently resuspend and dispense the contents of the vial into the poly-L-lysine-coated culture vessel. A seeding density of 10,000-50,000 cells/cm² is recommended, with an optimal range of 20,000-25,000 cells/cm².

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.

6. Replace the cap or lid of the culture vessel 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 best results, do not disturb the culture for at least 16 hours after the culture has been initiated. Refresh culture medium the next day to remove residual DMSO and unattached cells, then every other day thereafter.

Maintenance of 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 testes 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, 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).