Rat Pituitary Cells (RPC)

Catalog Number: R1200

Cell Specification

The pituitary is a small, pea-sized gland located at the base of the brain. It is responsible for controlling and coordinating: 1) growth and development; 2) the function of various body organs; and 3) the function of other glandular organs. The study of pituitary gland development provides a remarkable example of cell specification. There are six major cell types of the anterior (endocrine) pituitary, and eight hormones: growth hormone, prolactin, thyrotrophin stimulating hormone, adrenocorticotrophic hormone, leutinizing hormone, follicle stimulating hormone, melanocyte stimulating hormone, and endorphin are produced by different pituitary cells, which are located in specific regions of the pituitary gland. Many pituitary cell types have been shown to express natriuretic peptide receptors and to respond to natriuretic peptides, to stimulate cGMP accumulation [1].

RPC from ScienCell Research Laboratories are isolated from neonate day 8 rats and cryopreserved in primary culture with further purification and expansion. Each vial contains >5 x 10^5 cells in 1 ml volume. RPC are negative for mycoplasma, bacteria, yeast and fungi. RPC 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 RPC *in vitro*.

Product Use

RPC 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] Fowkes, RC, Forrest-Owen, W, and McArdle, CA (2000) C-type natriuretic peptide (CNP) effects in anterior pituitary cell lines: evidence for homologous desensitisation of CNP-stimulated cGMP accumulation in alpha T3-1 gonadotroph-derived cells Instruction for culturing cells. *Journal of Endocrinology* 166, 195–203.

Instruction for culturing cells

Caution: Cryopreserved cells are very delicate. Thaw the vial in a 37^{o}C waterbath and return them to culture as quickly as possible with minimal handling!

Set up culture after receiving the ordering:

1. Place the vial in a 37^{o}C waterbath, hold and rotate the vial gently until the contents are completely thawed. Remove the vial from the waterbath immediately, wipe it dry, and transfer it to a sterile field. Rinse the vial with 70% ethanol, and then wipe to remove excess. Remove the cap, being careful not to touch the interior threads with fingers. Using 1 ml eppendorf pipet gently resuspends the contents of the vial.
2. Dispense the contents of the vial into the equilibrated, poly-L-lysine coated culture vessels. A seeding density of 7,500 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 DMSO residue in the culture. It is also important that rat pituitary cells are plated in poly-L-lysine coated culture vessels that promote cell attachment and growth.
3. Replace the cap or cover, and gently rock the vessel to distribute the cells evenly. Loosen caps if necessary to permit gas exchange.
4. Return the culture vessels to the incubator.
5. Change the growth medium 4 - 6 hours after plating to remove the residual DMSO and unattached cells, then every other day thereafter. A health culture will display normal cell morphology, nongranular cytoplasm, and the cell number will be doubled after two to three days in culture.

Subculture:

1. Subculture the cells when they are 90% confluent.
2. Prepare poly-L-lysine-coated cell culture flasks.
3. Warm medium, trypsin/EDTA solution, trypsin neutralization solution, and DPBS to room temperature.
4. Rinse the cells with DPBS.
5. Incubate cells with 3 ml of trypsin/EDTA solution (in the case of T-25 flask) until 80% of cells are rounded up (monitored with microscope). Add 3 ml of trypsin neutralization solution to the digestion immediately and gently rock the culture vessel.

Note: Use ScienCell Research Laboratories' trypsin/EDTA solution that is optimized to minimize the killing of the cells by over trypsinization.

6. Harvest and transfer released cells into a 15 ml centrifuge tube. Rinse the flask with another 3 ml of growth medium to collect the residue cells. Examine the flask under microscope to make sure the harvesting is successful by looking at the number of cells left behind. There should be less than 5%.
7. Centrifuge the harvested cell suspension at 1000 rpm for 5 min and resuspend cells in growth medium.
8. Count cells and plate them in a new, poly-L-lysine-coated flask with cell density as recommended.

Caution: Handling human derived products is potentially bioharzadous. Although each cell strain testes negative for HIV, HBV and HCV DNA, proper precautions mush 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).