# Rat Microglia (RM)

Catalog Number: R1900

## Cell Specification

Microglia, one of the glial cell types in the CNS, is an important integral component of the neuro-glial cell network [1]. They have been observed in the brain parenchyma from the early stage of development to the mature state. Microglia act as brain macrophages when programmed cell death occurs during brain development or when the CNS is injured or pathologically damaged. Microglia can be considered the main cell in brain immune surveillance, can present antigens in the molecular context of MHC class II expression to CD-4 positive T cells, are capable of Fc-mediated phagocytosis, and share many common antigens with hemopoietic and tissue macrophages [2]. Furthermore, there is accumulating evidence that microglia are involved in a variety of physiological and pathological processes in the brain by interacting with neurons and other glial cells and through production of biologically active substances such as growth factors, cytokines, and other factors [3].

RM from ScienCell Research Laboratories is isolated from day 2 rat brain tissue. Cells are harvested after purification and delivered frozen. Each vial contains >1 x 10^6 cells in 1 ml volume. RM is characterized by immunofluorescent method with antibody to OX-42 (CD 11b/c). RM is negative mycoplasma, bacteria, yeast and fungi. RM are not recommended for long term culture since microglia do not proliferate in regular culture.

## Product Use

RM is for research use only. They are not approved for human or animal use, or for application in *in vitro* diagnostic procedures.

## Recommended Medium

It is recommended to use Microglia Medium (MM, Cat. No. 1901) for the culturing of RM *in vitro*.

## Storage

Transfer frozen 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 or gel ice.

## Reference


[1] Lee, S. C., Liu, W., Brosnan, C. F. and Dickson, D. W. (1992) Characterization of primary human fetal dissociated central nervous system cultures with an emphasis on microgia. Laboratory Investigation. 67:465-476.

[2] Fedoroff, S., Zhai, R. and Novak, J. P. (1997) Microglia and astroglia have a common progenitor cell. J. Neurosci. Res. 50: 477-486.

[3] Stoll, G. and Jander, S. (1999) The role of microglia and macrophages in the pathophysiology of the CNS. Prog. Neurobiol. 58:233-247.

# 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 order:**

1. Prepare a poly-L-lysine coated flask (2 μg/cm$^2$, T-25 flask is recommended). Add 5 ml of sterile water to a T-25 flask and then add 5 μl of poly-L-lysine stock solution (10 mg/ml, ScienCell cat. no. 0413). Leave the flask in incubator overnight (minimum one hour at 37$^{o}$C incubator).
2. 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.
3. Rinse the poly-L-lysine coated flask with sterile water twice and add 7 ml of complete medium to the flask. Leave the flask in the hood and go to thaw the cells.
4. 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 pipette gently resuspend the contents of the vial.
5. Dispense the contents of the vial into the equilibrated, poly-L-lysine coated culture vessels. A seeding density of ≥10,000 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 DMSO residue in the culture. It is also important that cells are plated in poly-L-lysine coated culture vessels that promote the cell attachment.*
6. Replace the cap or cover, and gently rock the vessel to distribute the cells evenly. Loosen caps if necessary to permit gas exchange.
7. Return the culture vessels to the incubator.
8. 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. A health culture of microglia show characteristic elongated, almost bipolar cell bodies with spine-like processes that often branch perpendicularly.

**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 microglia 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 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).