



## Human Thymic Epithelial Cells (HTyEpiC) Catalog #3910

### Cell Specification

Thymic epithelial cells (TyEpiC) are vital regulators of thymocyte development and T lymphocyte (T-cell) tolerance [1]. The thymus contains two types of TyEpiC, cortical and medullary thymic epithelial cells, which regulate the positive selection of thymocytes and the negative selection of autoreactive T-cells [2]. The inability of TyEpiC to remove autoreactive T-cells in the thymus can affect self-tolerance and contribute to the development of autoimmune diseases such as myasthenia gravis, type 1 diabetes, rheumatoid arthritis, and multiple sclerosis [3]. Studies have shown that changes in the expression of tumor necrosis factor receptor family members and transcription factors Foxn1 and autoimmune regulator (Aire) can alter the thymic microenvironment and self-tolerance [3]. Human TyEpiC are an excellent *in vitro* model to study the mechanisms that modulate TyEpiC functionality and to develop targeted treatments for autoimmune disorders.

HTyEpiC from ScienCell Research Laboratories are isolated from human thymus. HTyEpiC are cryopreserved at passage one and delivered frozen. Each vial contains  $>5 \times 10^5$  cells in 1 ml volume. HTyEpiC are characterized by immunofluorescence with antibodies specific to cytokeratin-18 and/or cytokeratin-19. HTyEpiC are negative for HIV-1, HBV, HCV, mycoplasma, bacteria, yeast and fungi. HTyEpiC are guaranteed to further culture under the conditions provided by ScienCell Research Laboratories; however, *HTyEpiC are not recommended for expanding or long-term cultures due to limited expansion capacity.*

### Recommended Medium

It is recommended to use Thymic Epithelial Cell Medium (TyEpiCM, Cat. #3911) for culturing HTyEpiC *in vitro*.

### Product Use

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

### Storage

Upon receiving, directly and immediately transfer the cells from dry ice to liquid nitrogen and keep the cells in liquid nitrogen until they are needed for experiments.

### Shipping

Dry ice.

### References

- [1] Alexandropoulos K, Danzl N. (2012) "Thymic epithelial cells: antigen presenting cells that regulate T-cell repertoire and tolerance development." *Immunol Res.* 54(1-3): 177-190.
- [2] Danzl N, Jeong S, Choi Y, Alexandropoulos K. (2014) "Identification of novel thymic epithelial cell subsets whose differentiation is regulated by RANKL and Traf6." *PLoS One.* 9(1): e86129.
- [3] Sun L, Hongran L, Luo H, Zhao Y. (2014) "Thymic epithelial cell development and its dysfunction in human diseases." *Biomed Res Int.* 2014: 206929.

## **Instructions for culturing primary cells**

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**Caution:** Cryopreserved primary 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! Do not centrifuge the cells after thawing as this can damage the cells.

### **Initiating the culture:**

**Note:** ScienCell primary cells must be cultured in a 37°C, 5% CO<sub>2</sub> incubator. Cells are only warranted if ScienCell media and reagents are used and the recommended protocols are followed.

1. Prepare a poly-L-lysine-coated culture vessel (2 µg/cm<sup>2</sup>, T-75 flask is recommended). To obtain a 2 µg/cm<sup>2</sup> poly-L-lysine-coated culture vessel, add 10 ml of sterile water to a T-75 flask and then add 15 µl of poly-L-lysine stock solution (10 mg/ml, Cat. #0413). Leave the vessel in a 37°C incubator overnight (or for a minimum of one hour).
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. Rinse the poly-L-lysine-coated vessel twice with sterile water and then add 20 ml of complete medium. 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.
5. Carefully remove the cap without touching the interior threads. Gently resuspend and dispense the contents of the vial into the equilibrated, poly-L-lysine-coated culture vessel. A seeding density of 10,000-12,000 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 residual DMSO in the culture. It is also important that cells are plated in poly-L-lysine-coated culture vessels to promote cell attachment.***

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. Do not disturb the culture for at least 16 hours after initiation. Refresh culture medium the next day to remove residual DMSO and unattached cells.

### **Maintaining the culture:**

1. Refresh supplemented culture medium the next morning after establishing a culture from cryopreserved cells.

2. Change the medium every three days thereafter. Use promptly for experiments.

**Note:** We do not recommend cryopreservation of primary cells by the end user. Refreezing cells may damage them and affect cell performance. ScienCell does not guarantee primary cells cryopreserved by the end user.

*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 with 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.