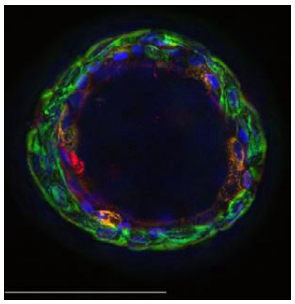
Extraction of Three-Dimensional Structures from Corning® Matrigel® Matrix

Guidelines for Use



Representative photomicrograph of 23-day-old human airway organoid that was extracted from Corning Matrigel matrix and then fluorescently labeled with specific markers. Basal cells (green), ciliated cells (red), mucus production from goblet cells (orange), nuclei (blue). Image captured using 40X objective. Scale bar is 100 µm.

Introduction

Corning Matrigel matrix is a commonly utilized tool for creating three-dimensional (3D) spheroids and organoids. Often the recovery of the 3D structures from the matrix is required for downstream applications such as DNA extraction, histology, or immunostaining. Here we provide guidelines for recovering 3D structures that are embedded in Matrigel matrix. The Matrigel matrix concentration, culture length, and size of the 3D structures will have an impact on the effectiveness of this protocol. Some optimization of volume, wash steps, and incubation time may be required.

Materials

- Three-dimensional structures embedded in Corning Matrigel matrix
- Axygen® wide bore tips (Corning Cat. No. TF-205-WB-R-S)
- Phosphate buffered saline (PBS; Corning Cat. No. 21-040-CV)
- Corning Cell Recovery solution (Corning Cat. No. 354253)

Procedure

1. Remove as much cell culture medium from the culture as possible taking care not to disturb the cells.
2. Add pre-chilled Corning Cell Recovery solution at a volume ≥2X that of the Matrigel matrix volume.
3. Pipette up and down gently using wide bore tips to carefully break up the Matrigel matrix without damaging the 3D cultures.
4. Incubate cultures with Corning Cell Recovery solution at 4°C for approximately 20 minutes.
5. Visualize cultures under the microscope to see if the Matrigel matrix has been fully depolymerized and 3D cultures are floating free from Matrigel matrix. It may be necessary to briefly centrifuge cultures, remove the Cell Recovery solution, and repeat steps 2 through 5.
6. If 3D cultures appear free from Matrigel matrix, briefly centrifuge cultures to separate structures from the solution. Remove the Cell Recovery solution and wash cultures with cold PBS several times.

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