
## ◆ Cellrix® 3D Culture System (24 well kit)

**Cellrix® 3D Culture System**, an innovative 3D cell culture system based on alginate and gelatin, which is providing a physiological environment much closer to the biological reality allowing encapsulated cells to preserve the predisposed biochemistry and phenotype. Stable and uniform 3D shape can be formed with application of the casting mold technology with 2-stage double cross-linking and check the exact end point of 3D formation by monitoring the color change of casting gel.

**Cellrix® 3D Culture System** is compatible with all standard **analytical technologies**: Being transparent makes it suitable for **microscopy: brightfield, immunofluorescence.** Rapid and gentle method using the dissolving buffer to retrieve the cells is enables to the use of **flow cytometry**. And protein and nucleic acids can be extracted easily by retrieve the cells for **PCR, RT-PCR, Western Blot**.

**Cellrix® 3D Culture System** is suitable for research of Stem Cell / Spheroid 3D Culture and Cancer Research / Cell Signaling / Tissue Engineering / Regeneration / Cell Delivery, and gives a more predictive tool in various applications: drug development, toxicity assessment, cell-based assays in many different research areas such as cancer therapy or stem cell research.

**■ Kit component**

| Product name | | Usage | Amount | Cat no. |
|---|---|---|---|---|
| **Cellrix® 3D culture system 24 well kit** (Cat no. B1010-024) | Bio-Gel | Hydrogel base for 3D culture | 5 mL | B1001-005 |
| | Casting Gel | Components for 3D formation | 30 mL x 2 | B1012-030 |
| | Casting mold | | 1pk (12 set) | B1013-024 |
| | Dissolving buffer | Reagent for recovery of 3D cultured cells | 20 mL x 3 | B1004-020 |

※ Related Products
- Cellrix® Firming buffer 50 mL, Cat no. B1005-050
- Cellrix® Bio-Gel 10 mL, Cat no. B1001-010
- Cellrix® Dissolving buffer 50mL, Cat no. B1004-050

## Cellrix® 3D Culture System - 24well kit

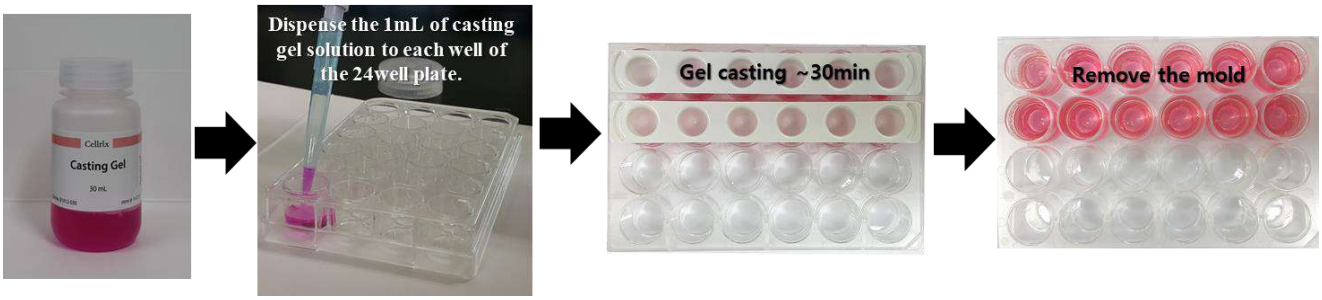
### 1. Preparation of casting gel

① Dissolve the the Cellrix® **C**asting gel at 70~80℃ (use the microwave oven)

② Dispense the 1mL of casting gel solution to each well of the 24 well plate.

③ Put the casting mold and cover the 24 well plate. After then stand at 4℃ for 30min.

④ Remove the casting mold.




### 2. Mix with the Bio-Gel and cell

① Pre-warm up the Cellrix® Bio-Gel at 37℃

② Harvest cells according to best cell culture practices.

③ Resuspend cells in minimum volume of culture medium at the appropriate cell concentration (or you may resuspend the cells with Bio-Gel solution directly).

*※ Cell densities tested within the hydrogel can be optimized based on the cell type or purpose of experiment. General cell densities are ranged from 1 x $10^6$ to $10^7$ cell/ml.*

④ Slowly add the volume of the cell stock solution into the Bio-Gel solution. Trying not to introduce bubbles, swirl carefully around the mixing vessel to mix the solutions together.

### 3. Cell mixed 3D gel casting

① Dispense 100~150uL Cell mixed Bio-Gel solution to each well of Casting gel (24-well plate).

② Put the casting gel on the 37℃ for 20min, after then the 4℃ for 10min. You can check on the complete gelation when the red color of casting gel changed with the yellow color.

③ Use the sterile forceps (or spatula) to scoop Biogel from the casting gel and place each Bio-Gel into a separate well of the 24well plate.

④ Add the 1mL of culture medium into the each well, and incubate the optimum culture conditions.

### 4. 3D Cultured Cell Recovery

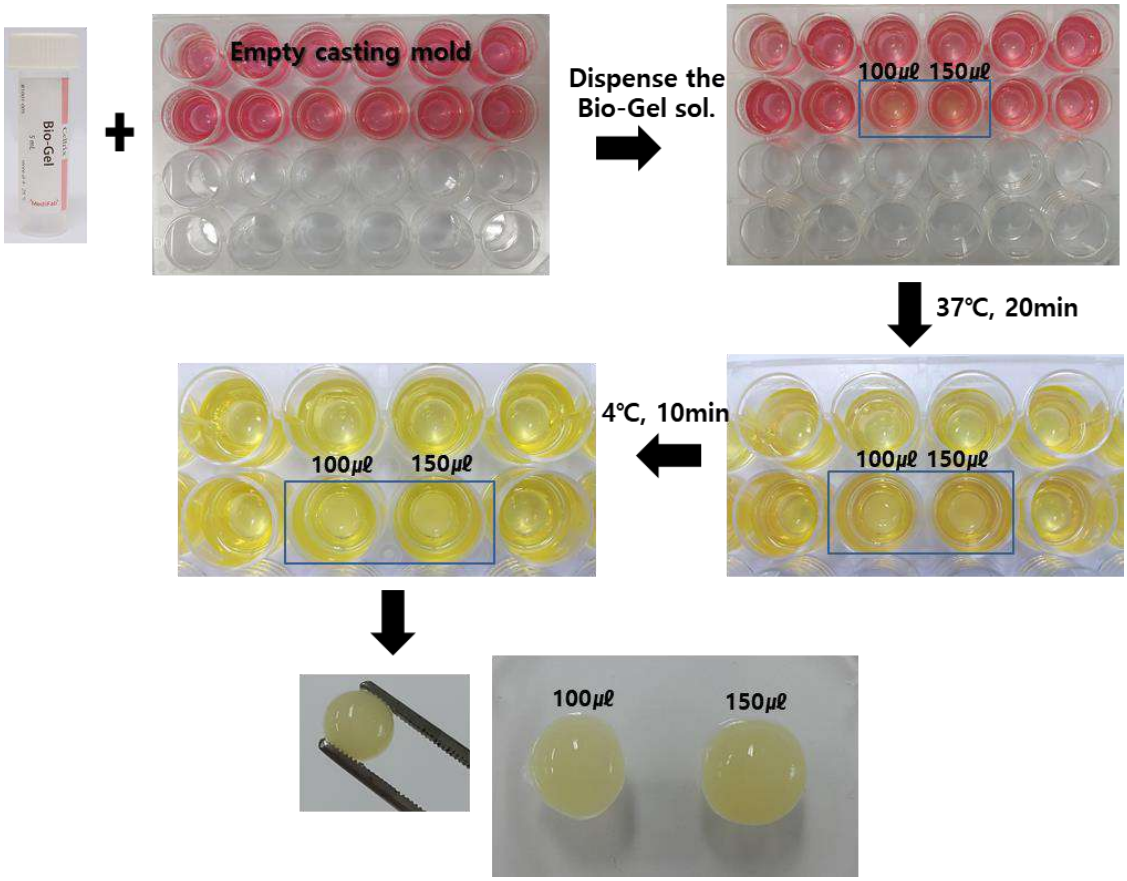
① Remove the medium from the entire well using an aspiration pipette.

② Add 0.5~1mL of dissolving buffer to each well, and incubate for 1 hour in the $CO_2$-incubator.

③ Transfer the dissolved solution to 1.5mL tube and centrifuge at approximately 300 ×g for 3 min.

⑤ Remove the supernatant, and wash with PBS buffer.

Option : If you want cell counting, add the trypan blue solution and count the cell with hemocytometer.




**Measuring Cell Viability – Recommend**

① Remove the medium from the entire well using an aspiration pipette

② Add 0.5 of dissolving buffer mixed with the 50 µL of Cellrix® Viability Assay kit to each well

③ Incubate for 0.5~ 4 hours at 37℃ in standard culture conditions.

④ Shake the plate briefly on a shaker and measure absorbance of treated and untreated cells using a plate reader at OD=450 nm.

**IMPORTANT NOTES**

- Bio-Gel concentration can be optimized based on the cell type mechanical needs.
- Cell densities tested within the hydrogel ranged from 1 x $10^6$ to 10 x $10^6$ cell/ml.
- Bio-Gel volume tested ranged from 100 µl to 200 µl. The use of positive displacement micropipettes ensures accuracy and precision when pipetting.
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