◆ Cellrix® 3D Culture System (96 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: bright field, 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 96 well kit (Cat no. B1000-096)	Bio-Gel	Hydrogel base for 3D culture	5 mL	B1001-005
	Casting Gel	Components for 3D formation	50 mL x 2	B1002-050
	Casting mold		1pk (12 set)	B1003-096
	Dissolving buffer	Reagent for recovery of 3D cultured cells	20 mL	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 - 96well kit

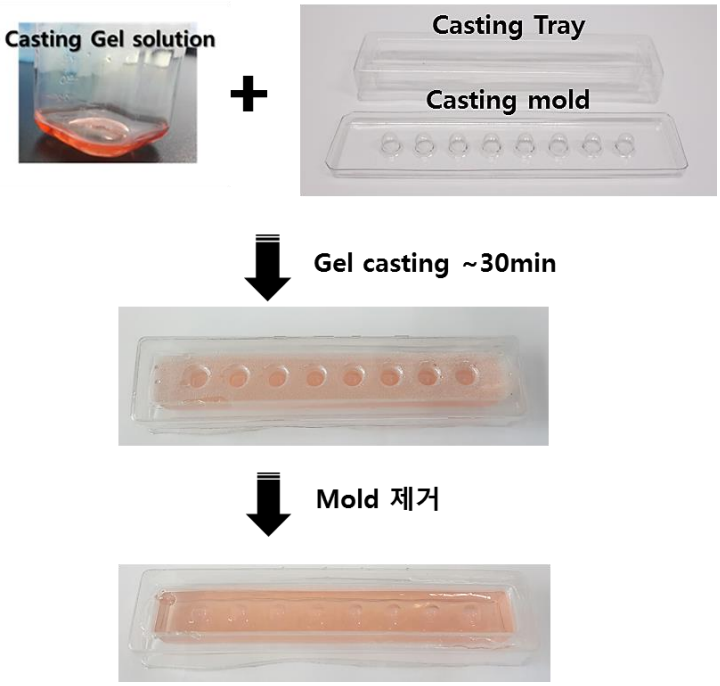
1. Preparation for casting gel

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

② Dispense the 5~6mL of casting gel solution to the casting tray.

③ Cover the casting tray with the mold, after then stand at 4℃ for 30min.

④ Remove the casting mold.



2. Mix 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.

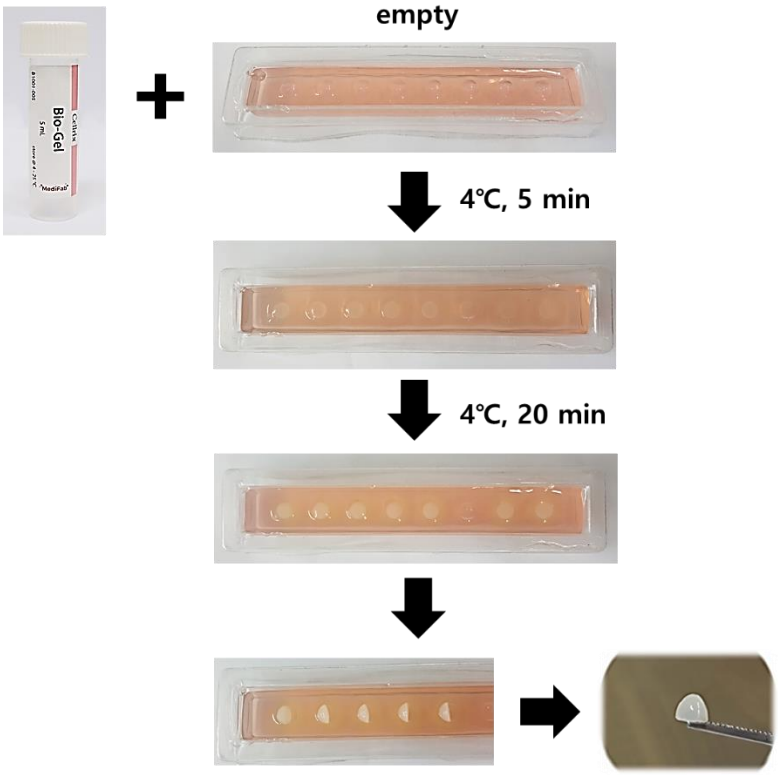
3. Cell mixed 3D gel casting

① Dispense 30~35uL cell mixed Bio-Gel solution to each well of casting gel (96-well plate test).

② Put the casting gel on the ice (or 4℃) for 20~30min. 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 Bio-Gel from the casting gel and place each Bio-Gel into a separate well of the 96-well plate.

④ Add the 200uL 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 100uL 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 DPBS 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 100uL of dissolving buffer to each well, and incubate for 1 hour in the CO_2-incubator.

③ Mix the 10 µL Cellrix® Viability Assay kit reagent and 100 µL dissolving buffer, and then add the 100 µL/well Cellrix® Viability Assay kit mixed solution into each well and 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 10^7 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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