



Biomaterials USA LLC

Provide Novel Biomaterials to the World

ShakeGel™ 3D
 3D Cell Culture Hydrogel

Storage:

The product is freshly manufactured. The product is sterilized. This product can be stored in room temperature (20-28 °C) for 6 months from the date of delivery.

DO NOT Freeze.

Product Description:

The ShakeGel™3D is bioactive hydrogel mimicking the microenvironment and microstructures of tissues in supporting 3D tissue-like growth *in vitro* and *in vivo*. This product is suitable for any cell type. We also provide customized functional hydrogels supporting proliferation or differentiation of stem cells.

Product Composition

Composition	Size	Condition
Hydrogel Solution	5 mL	Translucent/viscous
Lysis Buffer	15 mL	Clear thin fluid

Procedure

Note: It is recommended to pre-warm the hydrogel to 37°C before use.

1. Gently mix the gel solution and cell suspension into centrifuge tube. See Table 1 for recommended volumes. Then vortex slightly and quickly for 1 second. Repeat for 1-2 times.

Table 1: Recommended volumes

Plate size	Gel solution	Cell suspension	Culture medium
96-well plate	30µL/well	30µL/well	80µL/well
24-well plate	150µL/well	150µL/well	400µL/well
12-well plate	300µL/well	300µL/well	800µL/well
6-well plate	600µL/well	600µL/well	1600µL/well

Note: A) Mix well. B) Do not vortex the mixture more than 1 second each time neither more than three times. Excessive shaking would give rise to too many air bubbles in the gel. C) The recommended final concentration of cells is between 4.2×10^5 /mL ~ 1×10^6 /mL.



2. Pre-wet the cell culture wells with 1× Sterile PBS. Then, rapidly add the gel-cell mixture to the well and gently rock the culture-ware back and forth to spread the mixture evenly across the surface of the well. In addition, you can prepare microtissue by dropping the gel-cell mixture on any surface of sterile cell culture vessels directly.

Note: It is recommended to rinse the culture plate firstly since you have less than 20 minutes before the hydrogel forms.

3. Incubate at 37°C for 5-10minutes for hydrogel formation.
4. Add culture medium along the plate wall. See Table 1 for recommended volumes. Incubate at 37°C in a humidified atmosphere containing 5% CO₂.

Note: Do not shake the cell culture plate during this time to avoid the breaking the cell-gel constructs.

5. After 24 hours, discard half volume of old medium and add the same amount of fresh medium. After that, you can replace the culture medium routinely for the follow-up experiments.

Note: To avoid pipetting up cell-gel constructs, please gently pipet off old medium.

NOTES

- If you need to stain the cells, incubate the hydrogel with the dye.
- If you want to use the hydrogel in an animal model, you can mix the gel with cell suspension and inject to the animal.
- **Only** if you wish to retrieve the cells from the hydrogels, please operate according to the following steps.

Attention: The following steps will destroy the 3D structures of the cell-gel constructs.

1. Discard the culture medium. **Wash with lysis buffer.** Add lysis buffer. The volume ratio of lysis buffer to gel is 3:1. Pipette up and down thoroughly for 3-5 minutes at room temperature to completely lyse the hydrogel.
2. When the gel-cell mixture become entirely liquid, collect the mixture to a centrifuge tube. Rinse the plate with PBS for several times and collect the PBS to the above centrifuge tube. Then, centrifuge at 1500 rpm for 3 minutes.
3. Discard the supernatant and then harvest the cells.



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FAQs

Q1: How to avoid air bubble formation?

A1: Pre-wet pipet tips with PBS before use or alternatively, use a 1 ml sterile syringe without needle to aspirate the gel solution. Pipette the cell-hydrogel mixture up and down gently.

Q2: How to make the cell-hydrogel mixture spread evenly on the well surface?

A2: Pre-wet culture wells with 1×PBS before use. Add the cell-gel mixture along the plate wall and gently spread with a pipette.

Q3: What are the gelation conditions of the gel?

A3: Incubate at 37°C for 5-10 minutes for hydrogel formation.

Q4: What is the gray floc in the hydrogel?

A4: The gray floc is the bioactive ingredient in this product, which can promote cell growth.