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20240829

Porous Gelatin Methacryloyl

Porous GelMA

Product component

Item	character	Package Size	Notes
Porous GelMA hydrogel	White spongy	0.6 g/ bottle x2	Keep in dark

This instruction applies to EFL-GM-PR-001/002

Storage

Dry kit： room temperature, 3 months; 4℃, 12 months; -20℃, 18 months. **Sterile solution**：4°C (in dark), 7 days; -20℃ (in dark), 6 months. **Please note that repeated freezing and thawing of the solution will affect the performance of the product, so it is best to prepare it when using it.**

period of validity

The date of manufacture is shown in the package.

Required materials

- EFL-GM-PR series porous GelMA hydrogel products EFL
- EFL-LS-1601 series 405nm curing light source equipment EFL
- PBS（1X）
- Constant temperature magnetic stirring water baths
- 0.22 μm Sterile needle filters
- 10-50 mL Sterile centrifuge tubes
- 10 mL Syringes
- 1-5 mL Pipettes & tips

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Operation steps (3D cell culture)

Steps	Title	Materials	Processes
1	Prepare solution	➢ EFL-GM-PR series porous GelMA hydrogel products ➢ PBS（1X） ➢ Pipette guns ➢ constant temperature magnetic stirring water baths	1) Add appropriate amount of PBS to the porous GelMA bottle; Recommended concentration for porous GelMA is 6-8% w/v. (bottle contains magnetic rotor and 0.6g porous GelMA product). 2) Preparation of porous GelMA hydrogel precursor solution by **magnetic stirring at 37°C in a water bath keeping in dark for 1h (important step).**
2	Sterilise solution	➢ 0.22 μm Sterile needle filters ➢ Constant temperature water baths ➢ Sterile centrifuge tubes	1) Sterilise the above solution immediately with a sterile 0.22μm needle filter (to prevent gelation when the temperature drops) and store at 37°C in dark. Note: If it is not possible to use it all at once, store in the refrigerator for a short period of time (< 7 days). Before the next use, redissolve at 37°C and **shake for 20-30 seconds to homogenise the material.**
3	Mixe cells		1) Collecte cells. 2) Resuspension of cells with sterile precursor solution (multiple blowing or shaking).

<table>
<tr>
<td>4</td>
<td>Cure GelMA</td>
<td>➢ Pipettes & tips
➢ EFL-LS-1601 series 405nm curing light source equipment</td>
<td>1) Add precursor solution to the well plate; 96-well plate: 50-100 μL/ well, 48-well plate: 100-300 μL/ well, 24-well plate: 300-500 μL/ well.
2) Leave at room temperature for 2 min.
3) Irradiation with EFL-LS-1601 series 405nm light source to cure hydrogels, Irradiation times are shown in the table below：
<table>
<tr><th>Models</th><th>6%</th><th>7%</th><th>8%</th></tr>
<tr><td>001</td><td rowspan="2">16~18s</td><td rowspan="2">13~16s</td><td rowspan="2">8~10s</td></tr>
<tr><td>002</td></tr>
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<tr>
<td>5</td>
<td>Wash samples</td>
<td>➢ Pipettes & tips</td>
<td>1) Add medium and incubate at 37°C for 5 minutes.
2) Remove the medium.</td>
</tr>
<tr>
<td>6</td>
<td>Culture cell</td>
<td></td>
<td>1) Change medium, observe and photograph according to experimental design.</td>
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</table>

Operation steps (2D cell culture)

The main steps of 2D cell culture are the same as those of 3D culture, except that step 3 mixing cells. Once the sample has been washed in step 5, the cells are ready to be grown on the surface.