Directions for Use
PhotoGel®-RUT Methacrylated Gelatin Kit

GELATIN METHACRYLATE WITH RUTHENIUM KIT FOR PHOTOCROSSLINKABLE HYDROGELS

Catalog Number **#5273-1KIT**

Product Description

Three dimensional (3D) gels allow for the study of the effects of the mechanical properties of the extracellular matrix (ECM), such as density and rigidity, on cell development, migration, and morphology. Unlike 2D systems, 3D environments allow cell extensions to simultaneously interact with integrins on all cell surfaces, resulting in the activation of specific signaling pathways. Gel stiffness or rigidity also affects cell migration differently in 3D versus 2D environments.

Furthermore, integrin-independent mechanical interactions resulting from the entanglement of matrix fibrils with cell extensions are possible in 3D systems, but not in 2D systems where the cells are attached to a flat surface.

Advanced BioMatrix offers PhotoGel®, a purified gelatin methacrylate kit, which provides native-like 3D gelatin gels with the unique attributes to be prepared at various concentrations and photocrosslinked to provide various gel stiffness. Though it is denatured collagen, gelatin retains many natural binding motifs such as RGD and MMP sites.

The PhotoGel® RUT kit consists of gelatin methacrylate and a visible light photoinitiator.

Table 1:

Item	Catalog No.	Package Size
Methacrylated Gelatin, Lyophilized	5208	2 x 500 mg
Photoinitiator Ruthenium	5246-100MG	100 mg
Photoinitiator Sodium Persulfate	5247-500MG	500 mg

Gelatin Methacrylate is produced from porcine, type A, 300 bloom gelatin. Gelatin macromers containing primary amino groups were reacted with methacrylic anhydride (MA) to add methacrylate pendant groups.

Our Gelatin Methacrylate achieves a degree of substitution of >70% for maximum crosslinking and range of stiffness.

The photoinitiator solution consists of Ruthenium and Sodium Persulfate which needs to be formulated in 1X PBS or cell culture media, allowing for visible light photocrosslinking of the gelatin at 400-450 nm.

Characterization and Testing

The formulated PhotoGel® has the following characteristics as shown in Table 2.

Table 2:

Test	Specifications
Cell Compatibility	>70% viability
Grafting Efficiency	≥75%
Sterility	No growth

Storage/Stability:

The product ships on frozen gel packs. Upon receipt, store the gelatin methacrylate at -20°C. Store the ruthenium and sodium persulfate at room temperature.

Preparation Instructions

Note: Employ aseptic practices to maintain the sterility of the product throughout the preparation and handling of the collagen and other solutions.

Note: The following instructions are for a 10% gelatin methacrylate solution. Recommended concentrations are 5-20%

1. Warm 10 mL of sterile warm 1X PBS or 1X cell culture media to >60°C.

2. Add the 5 mL's of warmed solution to the amber vial containing 500 mg of lyophilized gelatin methacrylate.

3. Mix on a shaker table or rotator plate until fully solubilized. Keep warm (>37°C) if possible (eg. place your rotator in an incubator) to help with full solubilization.

4. Calculate the volume of photoinitiator to add by multiplying the volume of solubilized gelatin by 0.02. If the resulting number is 200 ul, for example, you will add 200 ul of ruthenium and 200 ul of sodium persulfate.

5. Solubilize the required amount of ruthenium (per step 4) at a concentration of 37.4 mg/ml in 1X PBS or cell culture media.

6. Solubilize the required amount of sodium persulfate (per step 4) at a concentration of 119 mg/ml in 1X PBS or cell culture media.

7. Add the ruthenium to the gelatin solution and fully mix until solution is homogeneous.

8. Add the sodium persulfate to the gelatin/ruthenium solution and mix until solution is homogeneous.

9. Add your cells to the gelatin/photoinitiator solution.

10. Dispense your gelatin/photoinitiator/cell solution into the desired dish (ie. 6-well plate, 48-well plate).

11. For photocrosslinking, place printed structure directly under a 400-450 nm visible light crosslinking source.

Any excess material can be refrigerated and stored. The material will gel. Warm back up to >30°C for it to become liquid again. We recommend only adding photoinitiator to the amount of gelatin to be used at that time.