# PhotoGel®-RUT, Methacrylated Gelatin Hydrogel Kit

3D CC Hydrogel Cat. # CC325

FOR RESEARCH USE ONLY. NOT FOR USE IN DIAGNOSTIC PROCEDURES. NOT FOR HUMAN OR ANIMAL CONSUMPTION. pack size: 1 Kit



**Data Sheet** 

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## Background

3D cell culture, including bioprinting, allows for the creation of more physiological cell models by allowing cells to simultaneously interact with integrins on all cell surfaces, resulting in the activation of specific signaling pathways not activated in traditional 2D cell culture methods. Hydrogels are water swollen polymers that allow for the culture of cells in 3dimensions and can have profound effects on cellular development, differentiation, migration, and function. New areas of tissue engineering such as 3D bioprinting, have utilized UV photocrosslinked methacrylated hydrogel biomaterials (PEGMA, GelMA, HAMA and ColMA etc.) to encapsulate cells to make printable bioinks.

The PhotoGel<sup>®</sup>-RUT, Methacrylated Gelatin Hydrogel Kit is based upon purified porcine gelatin methacrylate (GelMA), which when photocrosslinked provides a native-like 3D environment for cells. Gelatin derived from denatured collagen retains many natural cell binding motifs such as RGD and MMP sites. In addition to porcine gelatin methacrylate, the kit includes the photoinitiator ruthenium/sodium persulfate for users to easily fine tune their photocrosslinking experiments (i.e. altering hydrogel stiffness or gelling speeds). 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. The gelatin methacrylate achieves a degree of substitution of >70% for maximum crosslinking and range of stiffness.

### **Kit Components**

The PhotoGel®-RUT, Methacrylated Gelatin Hydrogel Kit (CC325) contains:

1) CC325-1 (Store at Room Temp): Ruthenium Photoinitiator, 1 X 100 mg (CS226450), Sodium Persulfate Photoinitiator, 1 X 500 mg (CS226449).

2) CC325-2 (Store at -20°C): Methacrylated Gelatin, 2 X 500 mg (CS226451).

### **Quality Control**

Appearance: Lyophilized powder Gelatin Content: ≥ 1 gram Sterility (USP modified): No Growth pH: 6.0-8.0 Grafting Efficiency: ≥ 75% Cell Compatibility: > 70% Cell Viability

#### References

1) Mikos, A, et al. A high-throughput approach to compare the biocompatibility of candidate bioink formulations. Bioprinting Volume 17, March 2020.



Figure 1. 3D printing of PhotoGel<sup>®</sup> Methacrylated Gelatin Hydrogels can be used as native bioinks for tissue engineering bioprinting applications.

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### Instructions for Use

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



**Figure 2. 3D culture of human cells using PhotoGel® hydrogels and ruthenium.** A) Human bone marrow mesenchymal stem cells (MSCs), B) Human MSCs and human vein endothelial cell (HUVEC) coculture and C) human fibroblasts were encapsulated in PhotoGel® hydrogels and crosslinked with ruthenium using a 400-450 nm visible light source.

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