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### **Study of PhotoGel**® **Thermal Gelation Kinetics**

#### **Highlights**

- **Concentration-Dependent Gelation**: Higher PhotoGel®-50 (GelMA, 50% degree of methacrylation) concentrations have faster gelation, narrower gelation temperature range, and yield greater solution viscosity and gel stiffness.
- **Impact of Cooling Rate:** A slower cooling rate (2 °C/min) enhances gelation speed, especially for higher concentration gels, while lower concentrations gel more slowly and at much lower temperatures.
- **Unique Processing Window**: PhotoGel<sup>®</sup> at 20 w/v% gels more rapidly within a narrower temperature range, whereas 10 and 5 w/v% PhotoGel<sup>®</sup> exhibit a broader gelation temperature window, providing users with greater flexibility in preparing cell-laden bioinks while achieving optimal viscosities.

#### **Recommendation For Bioprinting**

For extrusion bioprinting, we recommend the following steps:

(1) Prepare a 20 w/v% PhotoGe $\circledR$  stock solution by fully dissolving the lyophilized PhotoGe $\circledR$  at a high temperature (37–40  $^{\circ}$ C), then mix it with an equal volume of cell suspension to obtain a final working bioink concentration of 10 w/v% for printing.

(2) Set the syringe cartridge to approximately 25 °C (avoiding temperatures below 20 °C to prevent complete gelation) and incubate the bioink syringe in the cartridge for 5–10 minutes to achieve optimal viscosity before printing.

(3) Store unused stock solution at 4 °C and reheat to 37–40 °C for complete solubilization before mixing with the cells.



*\*Refer to [product Directions-for-Use](https://advancedbiomatrix.com/photogel-ink-50.html) for more information.*

**Figure 1. Stiffness changes of PhotoGel® 50% at 5 w/v%, 10 w/v%, and 20 w/v% measured by the ElastoSens Bio during the sol-gel transition through cooling and melting processes.** 

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#### **Introduction**

Thermal gelation of methacrylated gelatin (GelMA), as seen in PhotoGel® 50%, is pivotal for producing bioinks with controlled viscosity and stiffness, which are key factors in bioprinting accuracy and structural integrity. PhotoGel® exhibits reversible thermal gelation that enables precise control over gelation time and stiffness, supporting customized bioink formulations that meet the specific requirements of diverse tissue engineering applications. This white paper explores how factors like concentration and temperature ramping rates impact PhotoGel® gelation, providing a roadmap for optimizing bioink preparation in bioprinting.

#### **Results and Discussion**

To understand PhotoGel®'s gelation and melting behaviors, we first measured the gel stiffness (G') during temperature change using a contactless rheology measurement (Fig. 1). Higher concentrations like 20 w/v% showed significantly stiffer gels than 5 and 10 w/v% during cooling, with gelation starting earlier at higher concentrations. This rapid stiffness increase highlights the suitability of PhotoGel<sup>®</sup> for applications needing quick structural stability. Additionally, gelation took longer (~100 minutes) than melting (~50 minutes), making PhotoGel® an adaptable choice for various temperature-controlled environments.



**Figure 2. Viscosity change of PhotoGel® 50% at 5 w/v%, 10 w/v%, and 20 w/v% measured by a Bohlin viscometer during the sol-gel transition through cooling and melting processes at different rates (n = 2).** 

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A second test measured viscosity changes at cooling rates of 2 °C/min and 5 °C/min (Fig. 2). During cooling, higher concentrations (20% w/v) and slower rates (2°C/min) increased the gelation speed and yielded a faster viscosity change, as well as a higher gelation temperature. In contrast, the lower concentration (5% w/v PhotoGel®) remained low viscosity at higher temperatures and exhibited a delayed gelation compared to higher concentrations below 25°C. In the melting phase, higher concentrations retained viscosity longer, showing greater thermal stability, with similar melting patterns across ramp rates. Overall, the results suggested that concentration and cooling rate are key factors in controlling gelation and stability, useful for determining the optimal PhotoGel® concentration and cartridge temperature for bioink preparation in extrusion-based printing.

#### **Application Guide for Bioink Preparation**

For bioink preparation, customers can leverage PhotoGel®'s tunable gelation properties by adjusting concentration and cooling rates to achieve desired mechanical properties. Higher concentrations and slower cooling rates enhance viscosity and stability, making them ideal for complex, high-stability bioprinting. Conversely, lower concentrations with faster cooling can create more flexible, lower-viscosity bioinks suited to delicate, less rigid tissue structures. By fine-tuning these parameters, users can maximize PhotoGel®'s versatility for applications requiring customized gel stiffness, stability, and rapid gelation, which are all critical for advancing precision and efficiency in bioprinting.

#### **Materials and Methods**

PhotoGel<sup>®</sup> 50% DOM (Advanced Biomatrix, Cat #VL3500000050) was prepared according to the manufacturer's instructions and fully solubilized in Milli-Q water at three different concentrations: 5 w/v%, 10 w/v%, and 20 w/v%. Once solubilized, PhotoGel® was loaded into an Elastosens rheometer for contactless rheology measurements under a temperature sweep (Table 1).





In tandem, PhotoGel<sup>®</sup> samples were loaded onto a Bohlin rheometer (Bohlin Instrument Inc.) for contact rheology measurements at two temperature ramping rates (Table 2). It was essential to use a Bohlin rheometer for a more controlled external environment, and to ensure that the entirety of the sample was homogenously responding to changes in temperature.



#### **Table 2. Bohlin rheometer testing sequence**