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PhotoGel® 50% and 95% DoM Gelation Rheology

Abstract: *This study was conducted to assess and compare the rheological properties of the thermal gelation of PhotoGel® 50% and 95% degree of methacrylation (DoM) at different concentrations. PhotoGel® was incubated at 20°C for 60 minutes, allowing thermal gelation to occur, and the stiffness was measured against time. The results indicate that as the concentration of PhotoGel® increases, the maximum gel stiffness increases despite the degree of methacrylation. As expected, the PhotoGel® 50% DoM achieved higher thermal gelation stiffness overall compared to PhotoGel® 95%, since less of the original gelatin protein is modified and unavailable for thermal crosslinking. In addition, the higher the concentration, the quicker the onset of thermal gelation for both PhotoGel® 50% and 95% DoM.*

Materials

Name/Description	Part Number	Lot Number(s)
PhotoGel® 50% DoM	IKG125000005	8964
PhotoGel® 95% DoM	5208	8828, 8868, 9066
DPBS	21600-044 1x50 L	2814720

Results

Figures 1 and 2 below show the average stiffness curves of PhotoGel® 50% and PhotoGel® 95% DoM respectively at different concentrations. In both cases, as concentration increases, the maximum stiffness or shear storage modulus (G') also increases. Figure 3 shows the non-linear increasing trends in maximum G' for both PhotoGel® 50% and 95% with increasing concentration. While the increasing trend is similar for both, PhotoGel® 50% achieves a higher gelation stiffness compared to PhotoGel® 95% at equal concentrations.

Figures 4 and 5 depict the average tan δ values of each concentration for PhotoGel® 50% and 95%, respectively, where tan δ = Loss Modulus (G")/Storage Modulus (G'). Tan δ provides a measure of how elastic (tan $\delta < 1$) or plastic (tan $\delta > 1$) a viscoelastic material is; the curves in figures 4 and 5 show that PhotoGel® behaves mostly as an elastic material despite the degree of methacrylation. At equal concentrations, PhotoGel® 50% and 95% show similar peak tan δ values. The peaks represent the onset of transition from liquid to solid, where the higher the concentration, the quicker the onset of gelation for both PhotoGel® 50% and 95%.

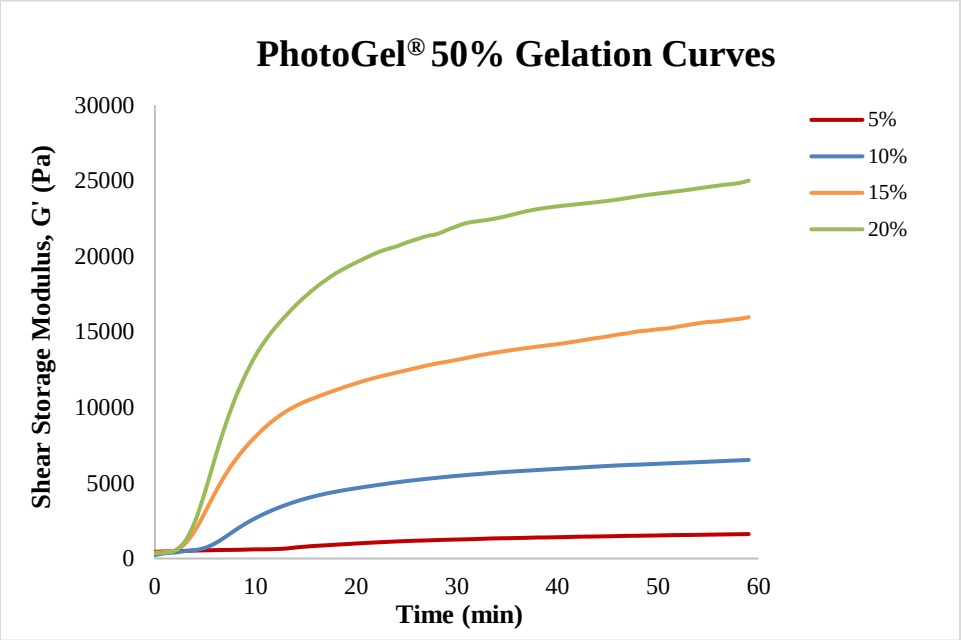

Figure 1. Stiffness curves due to thermal gelation of PhotoGel® 50% at varying concentrations.

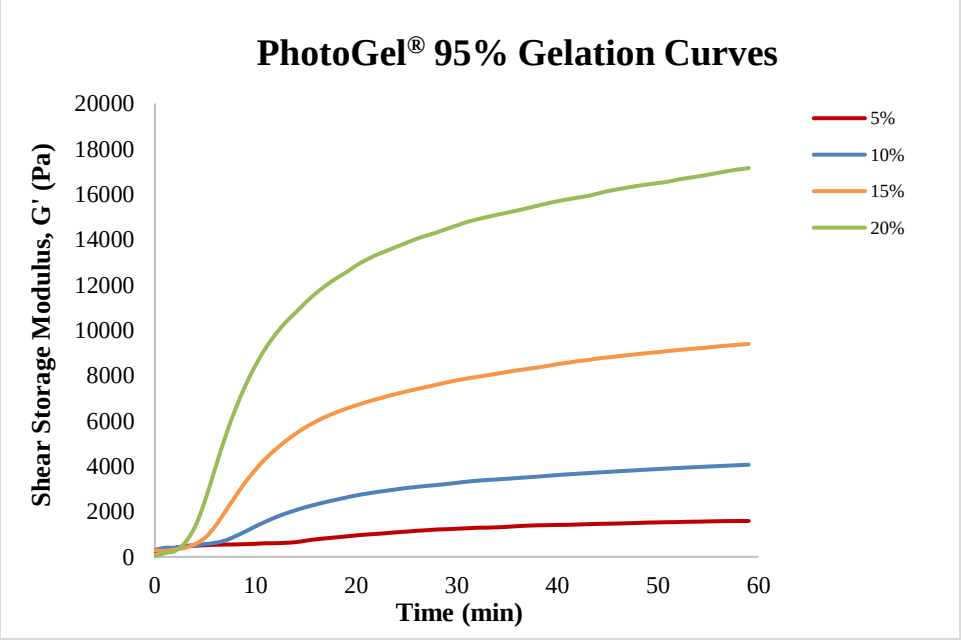

Figure 2. Stiffness curves due to thermal gelation of PhotoGel® 95% at varying concentrations.

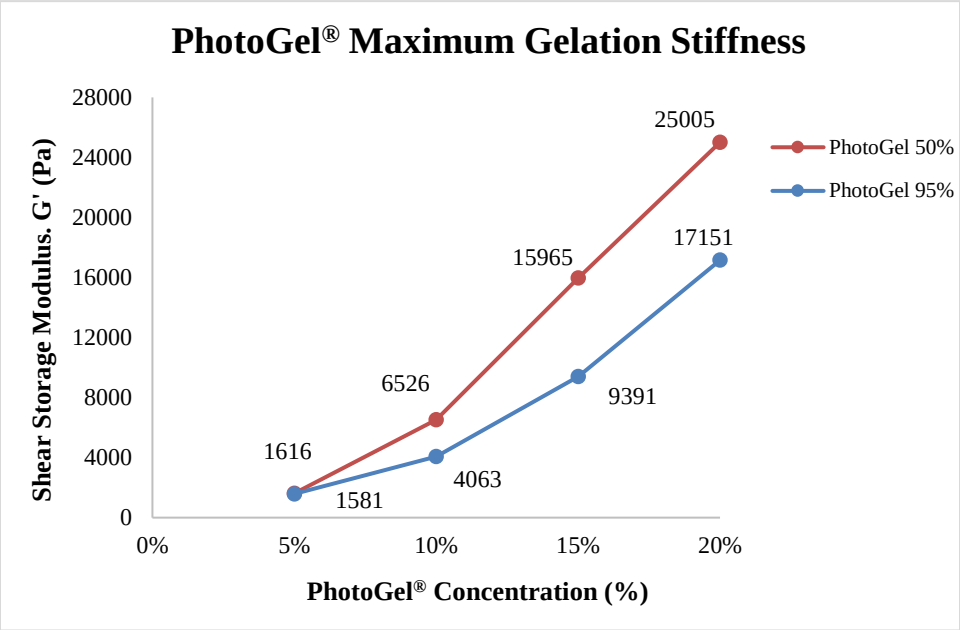

Figure 3. Increasing trend of maximum gelation stiffness vs. PhotoGel® concentration.

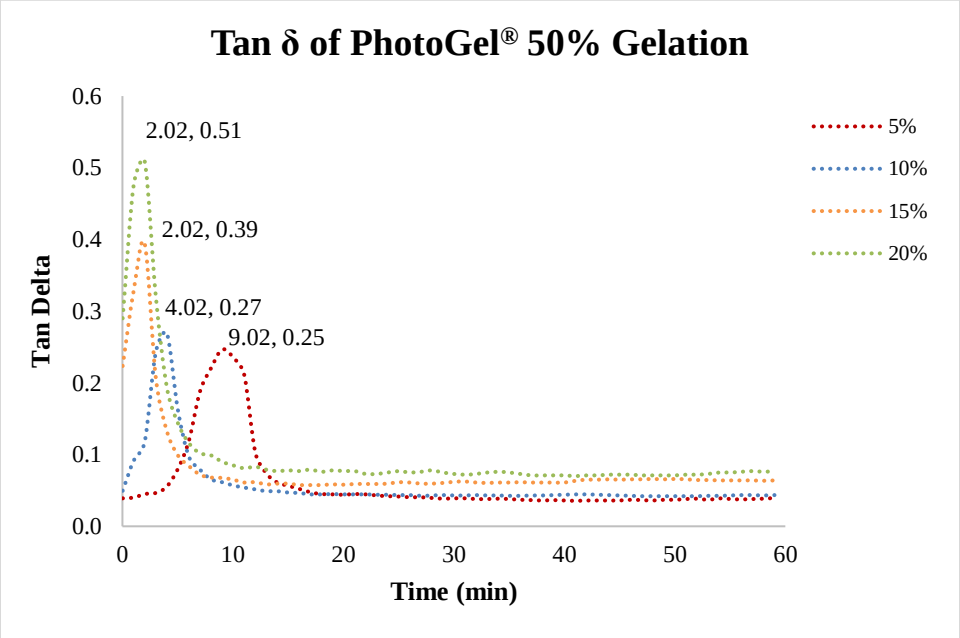

Figure 4. Tan δ curves of PhotoGel® 50% at different concentrations as gelation occurs.

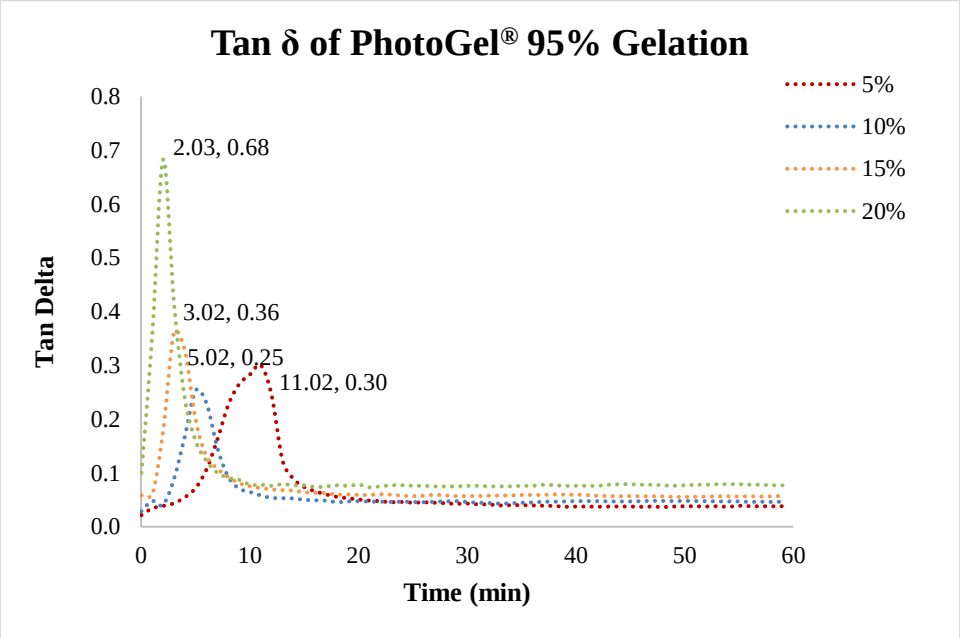

Figure 5. Tan δ curves of PhotoGel® 95% at different concentrations as gelation occurs.

Conclusion

In conclusion, it was found that for both PhotoGel® 50% and 95%, the higher the concentration, the quicker the onset of thermal gelation and the higher the maximum gel stiffness. Overall, PhotoGel® 50% achieved a higher thermal gelation stiffness compared to PhotoGel® 95%. PhotoGel® offers the unique capability to accurately fine tune hydrogel stiffness by dialing in various parameters such as concentration, degree of methacrylation, and gelation temperature, degree of photocrosslinking, etc. It is of special interest for bioprinting applications where varying the bioink properties such as concentration and degree of methacrylation, can significantly impact bioprinting performance, extrusion pressures, and hydrogel characteristics.

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Procedure

The following sample preparation and experimental set up procedures were performed to carry out the study. Briefly, a bulk solution of 20% PhotoGel® 50% DoM was diluted to 15%, 10% and 5% with 1X PBS, while lyophilized PhotoGel® 95% was reconstituted at 20% and diluted down with 1X PBS (see *"PhotoGel® Sample Preparations"* procedure below). Then, the Elastosens was calibrated, and the experimental parameters set (see *"Elastosens Experimental Set Up"* procedure below). Each PhotoGel® sample was then tested in duplicate sequentially and the test data collected and processed. The same bulk components, i.e. PhotoGel® 50%, 95% and 1X PBS, were used for all test groups in the study.

PhotoGel® Sample Preparations

1. The bulk 20% solution of PhotoGel® 50% was incubated at 40°C to liquefy.
2. The corresponding amount of 20% solution was aliquoted into individual tubes and kept at 40°C.
3. The corresponding amount of warm 1X PBS was added into each tube to yield concentrations of 5%, 10% and 15%.
4. 1X PBS was added into individual vials of lyophilized PhotoGel® 95% to yield a 20% concentration and incubated at 40°C on a shaker to mix gently until fully dissolved.
5. All solutions of 20% PhotoGel® were combined and further mixed for an hour to ensure homogeneity. The bulk solution was then aliquoted into individual tubes and kept at 40°C.
6. The corresponding amount of warm 1X PBS was added into each tube to yield concentrations of 5%, 10% and 15% and the solutions were mixed gently.
7. The solutions were covered in aluminum foil to minimize light exposure and incubated at 40°C prior to testing.

Elastosens Experimental Set Up

1. The Elastosens was turned on and calibrated once via vibration calibration.
2. The temperature was set to 20°C using the manual temperature control.
3. Once the temperature was equilibrated, a large empty sample cup was inserted in the testing chamber and secured. The same sample cup was used for all test groups in this study.
4. New sample files were created for each group in the following folders: PhotoGel 50% DOM > Gelation Curves and PhotoGel 95% DOM > Gelation Curves.
    a. File format: product name, test condition.
        i. E.g.: PhotoGel-50 5%.
        ii. E.g.: PhotoGel-95 10%.

5. The following test parameters were set for all test conditions:
    a. Type – Stiff.
    b. Sample File – Named as noted above.
    c. Test Name – Tester's initials followed by the number of the test run under that file name (each test was run in duplicate, and the resulting curves averaged).
        i. Ex - the first test ts01, second test ts02.
    d. Custom Information:
        i. Volume – 2 g.

ii. Oil – No.
iii. Concentration – Varies.
iv. Photoinitiator – N/A.
v. Photoinitiator concentration – N/A.
vi. Cup size – Large.
vii. Light intensity – N/A.
viii. Exposure time – N/A.
ix. Temperature – 20°C.

6. The following test sequences were set in the "Measurement Sequences" window:
    a. Sequence 1: Thermal incubation
        i. Duration: 60 min.
        ii. Step: 1 min.
        iii. Temperature configuration: manual.

7. The sample cup calibrated once prior to testing each group.
8. The sample cup was removed from the machine and placed on a scale to add 2 g ± 0.1 g of sample.
9. The sample-containing cup was placed into the machine and the test started.
10. After the first test was finished, a second test (following the procedure above) was run to obtain a duplicate test under the same file name.
11. The sample was removed from the cup after gelled, and the cup rinsed with milli-Q water and dried to be reused.