ACCSCIENCE PUBLISHING

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RESEARCH ARTICLE

Differentiation of iPSC-derived neural progenitors into motor neurons in 3D-printed bioscaffolds

Supplementary File

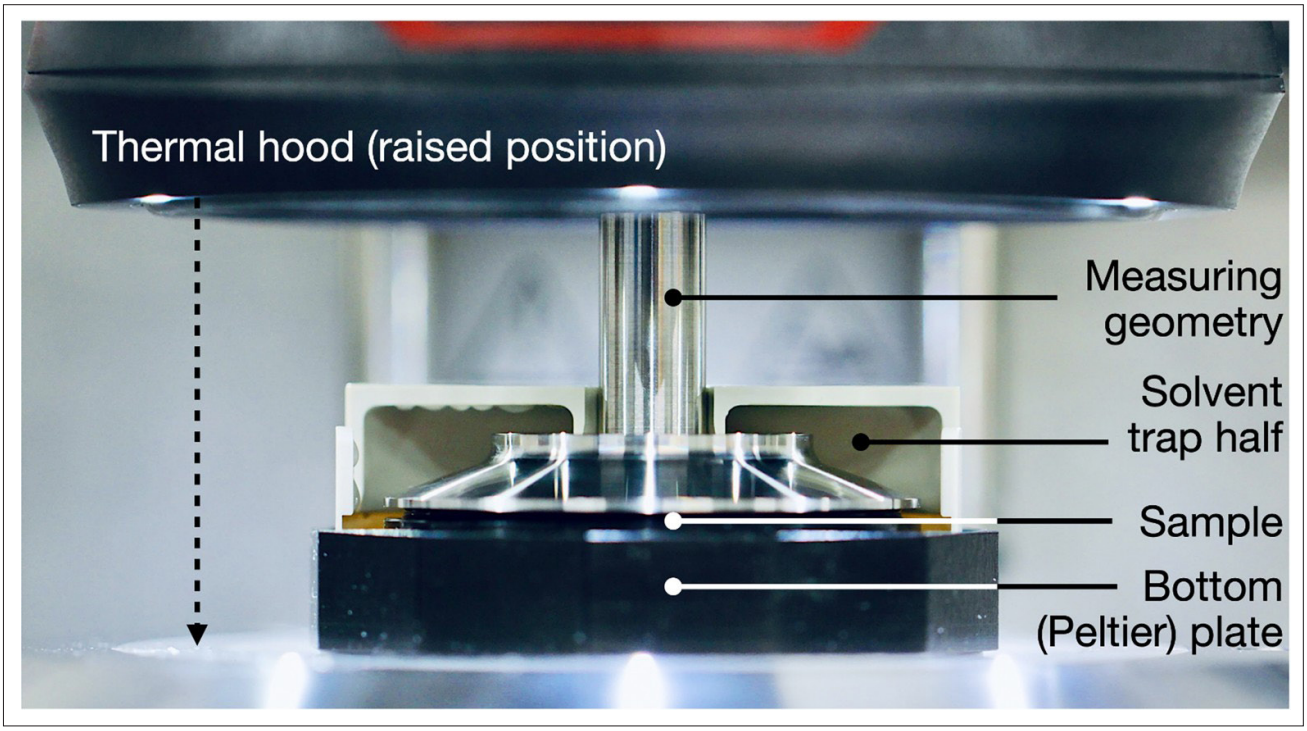


Figure S1. Overview of components in a rheological measuring setup.

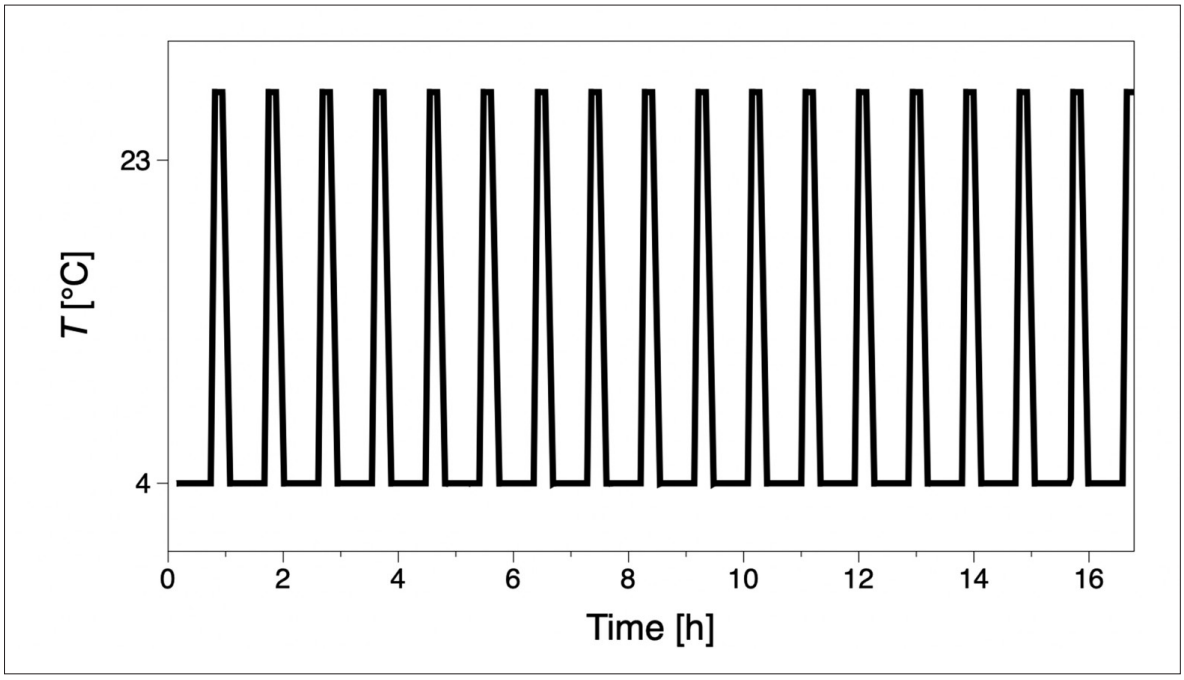


Figure S2. Illustration of the temperature oscillation test. Oscillatory shear measurements were performed at 4 and 27 °C.

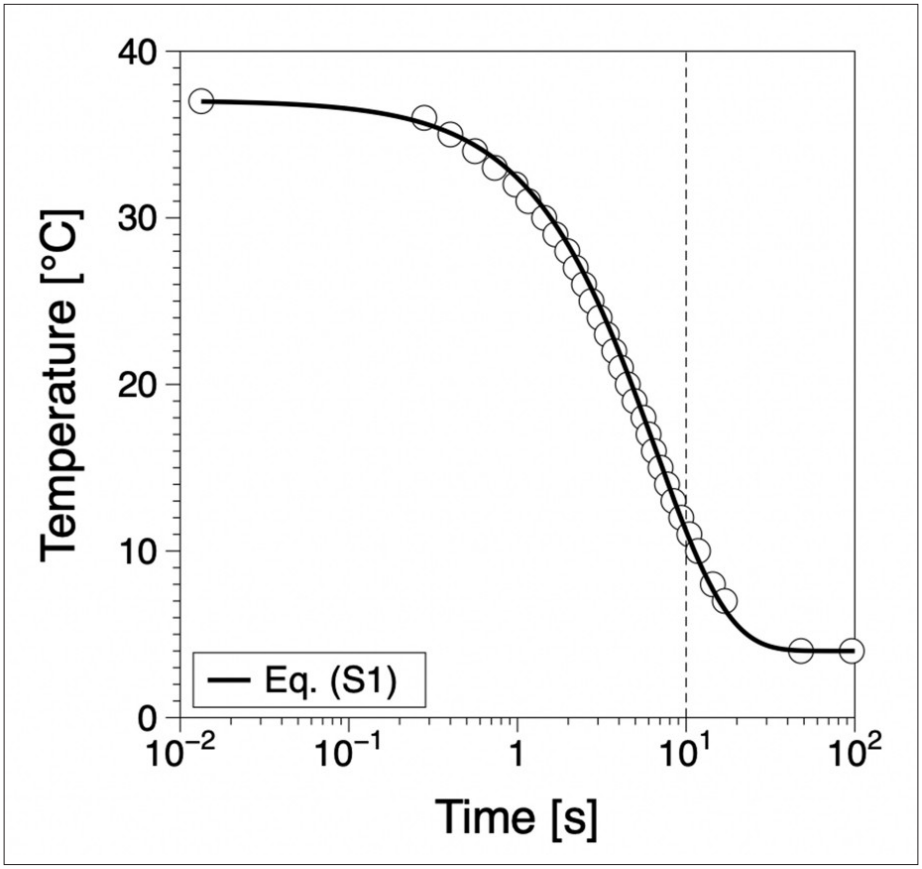


Figure S3. Measured cooling profile from the mixing temperature of 37 °C, after placing the 2 mL syringe containing 10% gelatin in a fridge with a temperature set at 4 °C. The solid line is a fit of **Equation (S1)**.

The rate of cooling can be approximated as being proportional to the temperature difference between an object and its' surrounding medium temperature using the Newton's cooling law. Thus, the temperature variation with time of the object is:

$$T(t) = T_{!} + \Delta T e^{"\#\$} \quad \text{(S1)}$$

where $\Delta T = (T_{\%} - T_{!})$ is the temperature difference between the initial temperature of the gelatin solution inside the syringe, $T_{\%}$, and the temperature of the ambient, $T_{!}$; and k is the heat transfer coefficient.

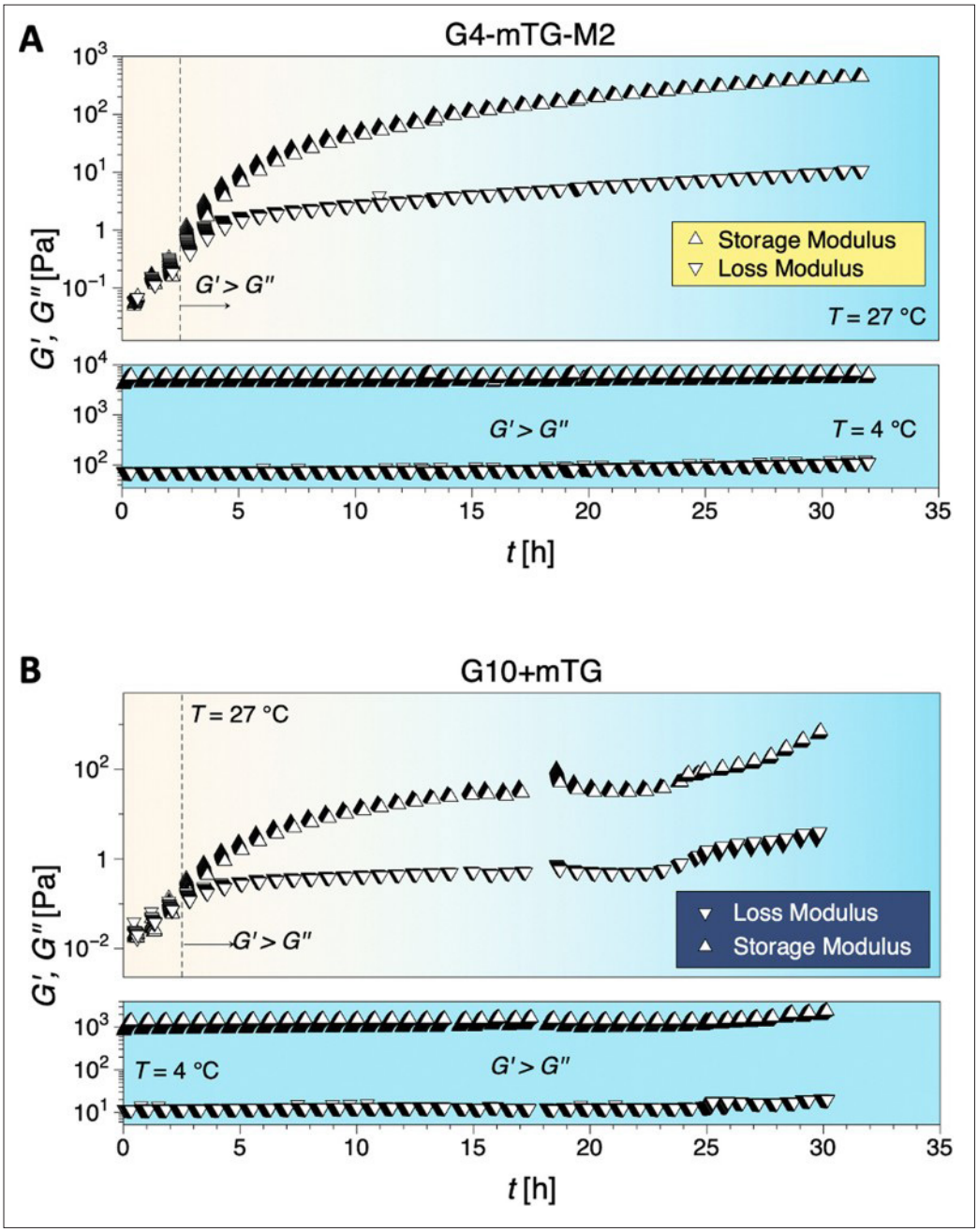
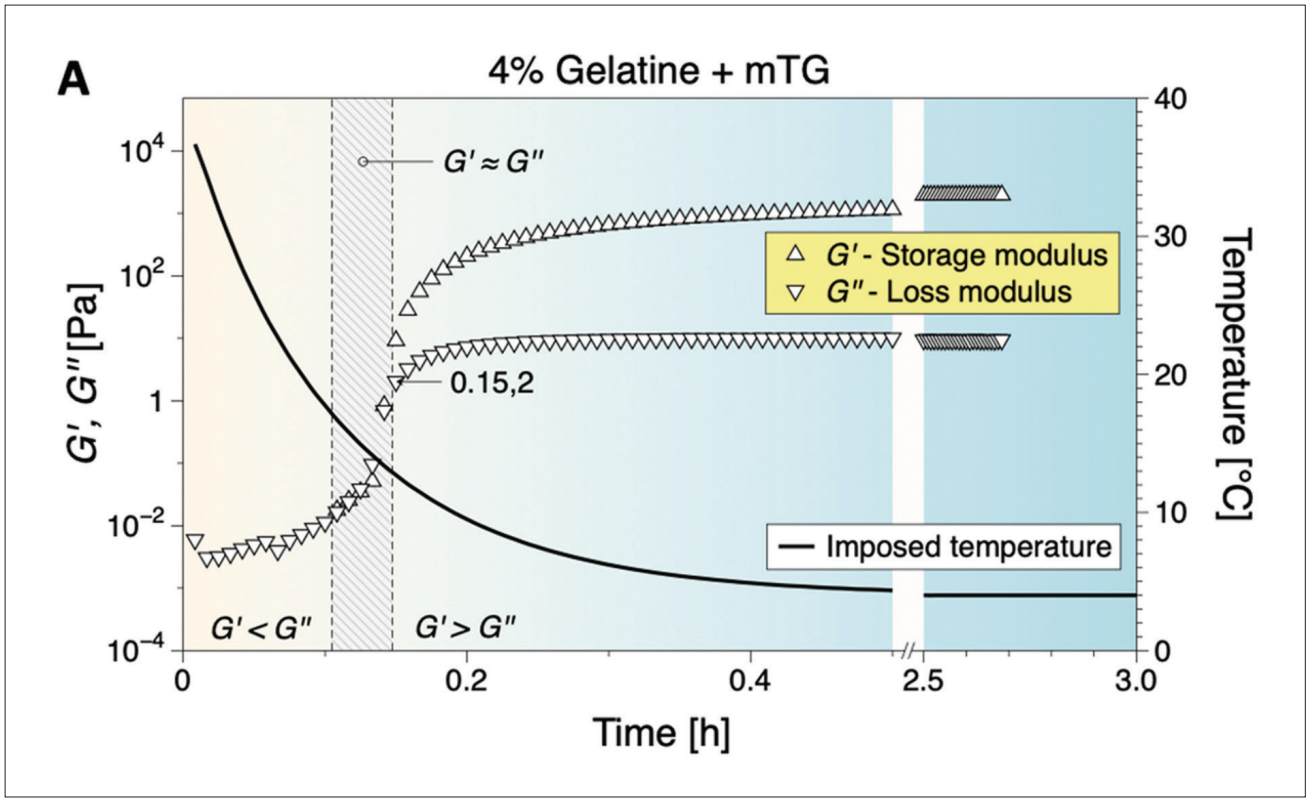
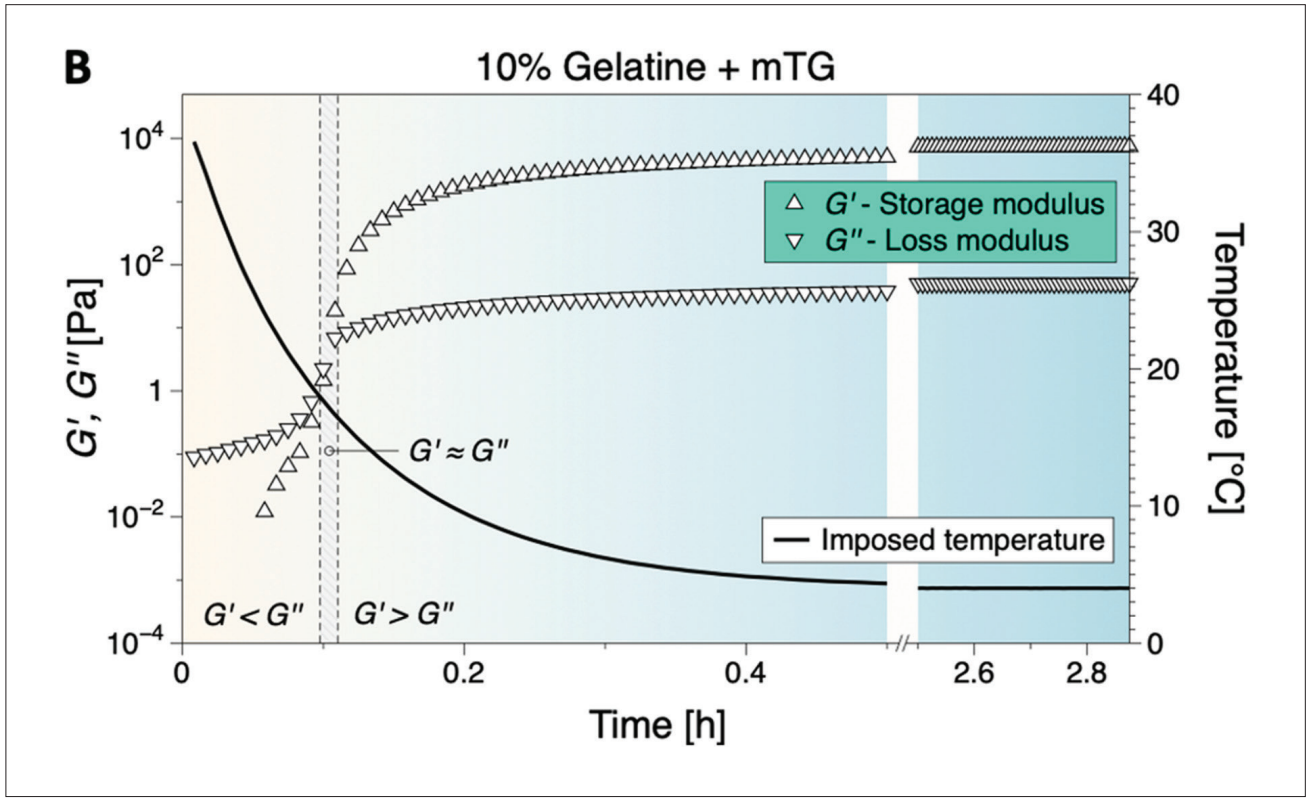
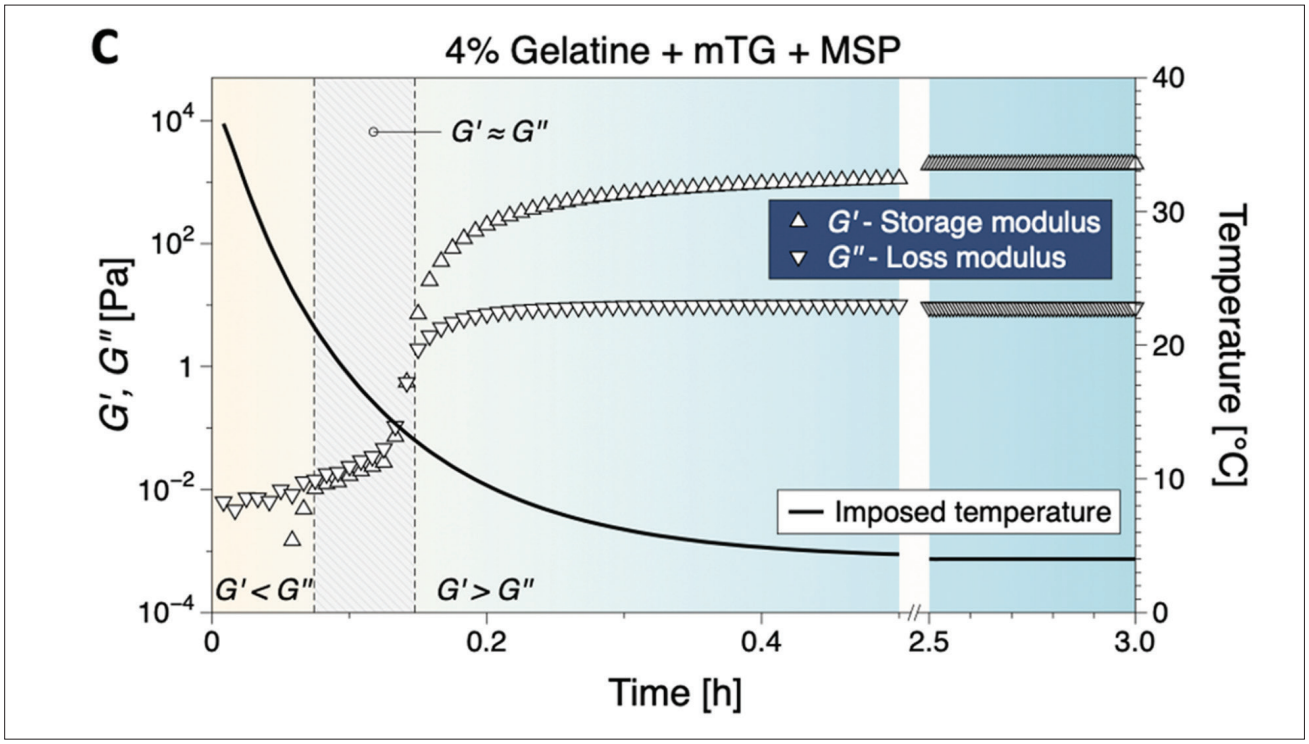


Figure S4. Temperature oscillation tests showing the evolution of the dynamic moduli with alternating temperature intervals between 4 and 27 °C with imposed temperature variation determined through Equation (S1) based on the cooling rate tests in Figure (S2).


A
4% Gelatine + mTG
G′ ≈ G″
G′ - Storage modulus
G″ - Loss modulus
0.15,2
Imposed temperature
G′ < G″
G′ > G″
G′, G″[Pa]
Temperature [°C]
Time [h]
10⁴
10²
1
10⁻²
10⁻⁴
40
30
20
10
0
0
0.2
0.4
2.5
3.0


B
10% Gelatine + mTG
G′ - Storage modulus
G″ - Loss modulus
G′ ≈ G″
Imposed temperature
G′ < G″
G′ > G″
G′, G″[Pa]
Temperature [°C]
Time [h]
10⁴
10²
1
10⁻²
10⁻⁴
40
30
20
10
0
0
0.2
0.4
2.6
2.8



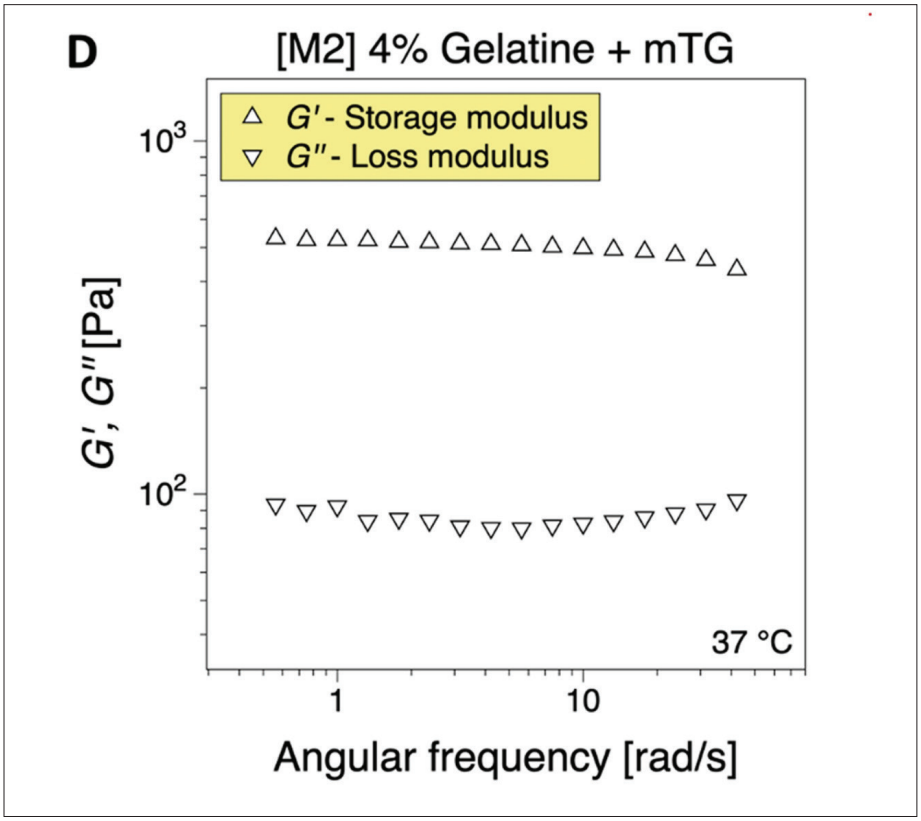


Figure S5. Cooling tests from 37 to 4 °C with imposed temperature variation through Equation (S1) (see data in Figure S2). Note that in Figure S2 the temperature is plotted on a logarithmic scale and that the data point density is lower compared to the data in this figure. The time required to reach a gel-like material response is summarized in Figure 1B. The frequency sweep in (*D*) was performed on a hydrogel prepared using Method 2 (M2) (outside the rheometer).

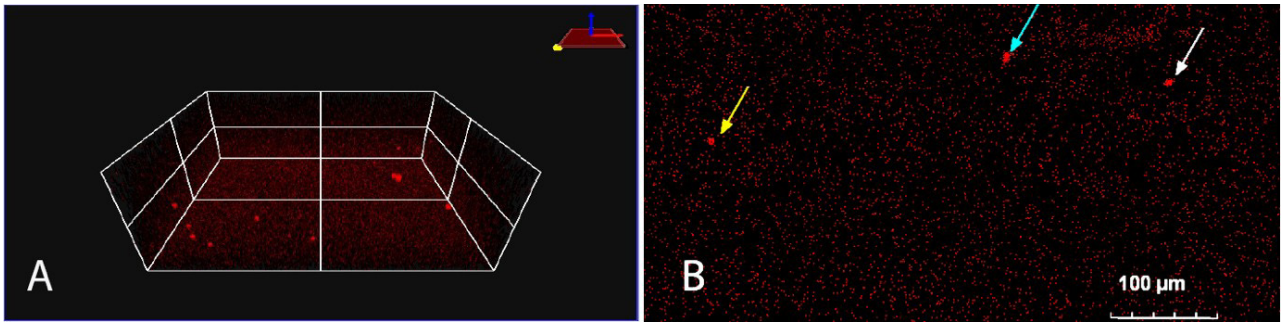


Figure S6. Confocal images of distribution of rhodamine-labeled MSPs inside the 3D-printed scaffold (A, B). Some MSPs were aggregated in small clusters (B). Abbreviation: MSPs, mesoporous silica particles.