Supporting Information

Multimaterial additive manufacturing of GelMA hydrogel-based structures with tuneable compositional and mechanical properties

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SI1. Resin formulations and characterization studies

Inks constituents were analysed by NMR (Figure S1) and formulation was systematically tunes to assess the effect of hydrogel concentration (from 10% to 30%) (Figure S2), photoabsorber concentration (Tartrazine concentration from 0.01 to 0.05%) and photoinitiator concentration (1% LAP). To establish printability window, light power and exposure time were scanned from 7mW/cm^2 to 50mW/cm^2). Based on shape definition and print reproducibility, the formulation with 20% GelMA + 0.025% Tartrazine + 1% LAP and printing parameters of exposure 2 sec, P = 10%=14 were selected for pμSLA printer. Printing resolution was found to be limited by in-built slicing software, hence slicing was refined (Figure S3). Degree of swelling of analysed (Figure S4).

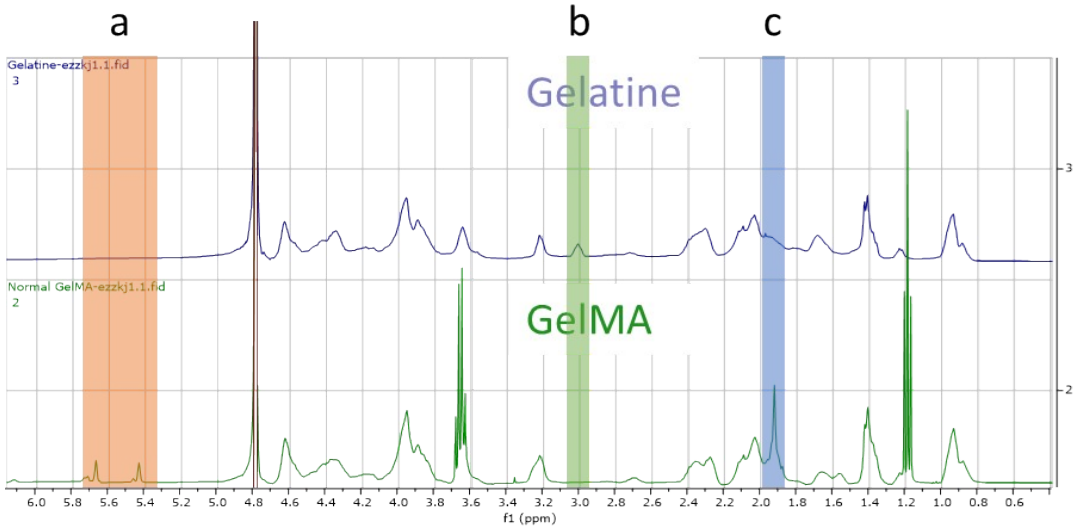

Figure S1. [1]H-NMR spectra of Gelatin and GelMA, where (a) confirms the presence of methacrylate group after gelatin functionalization. (b) confirms the formation of the amide bond, hence the shift of the peaks relative to the protons in the lysine groups, and (c) confirms the presence of the methacrylic groups.

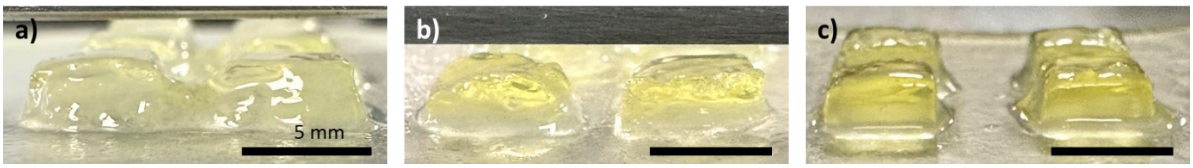

Figure S2. Photographs of GelMA hydrogels printed with different concentrations of GelMA **(a)** 10 w/v%, **(b)** 20 w/v%, **(c)** 30 w/v%.

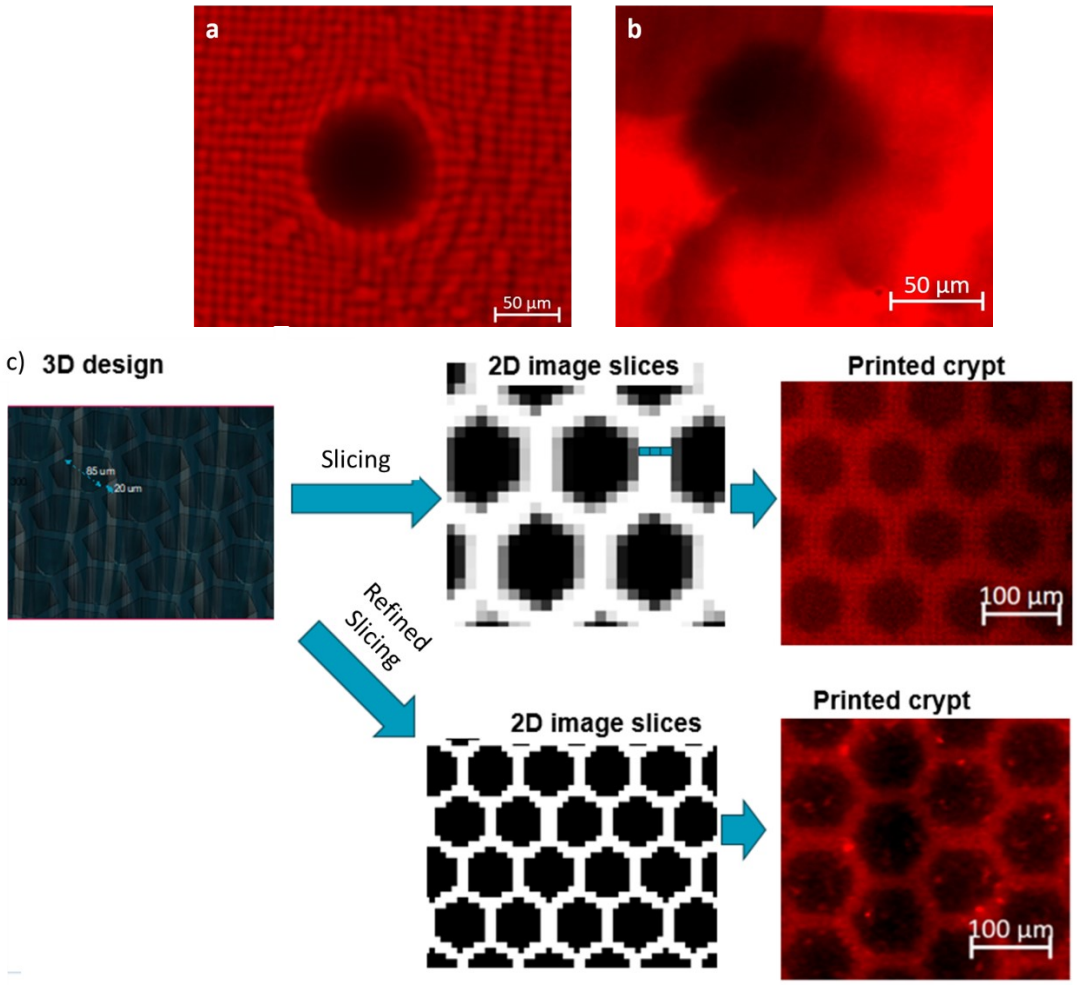

Figure S3. Fluorescent microscopy images of **(a)** top and **(b)** bottom slices of printed cavities. **(c)** Slicing refinement to achieve high definition printing.

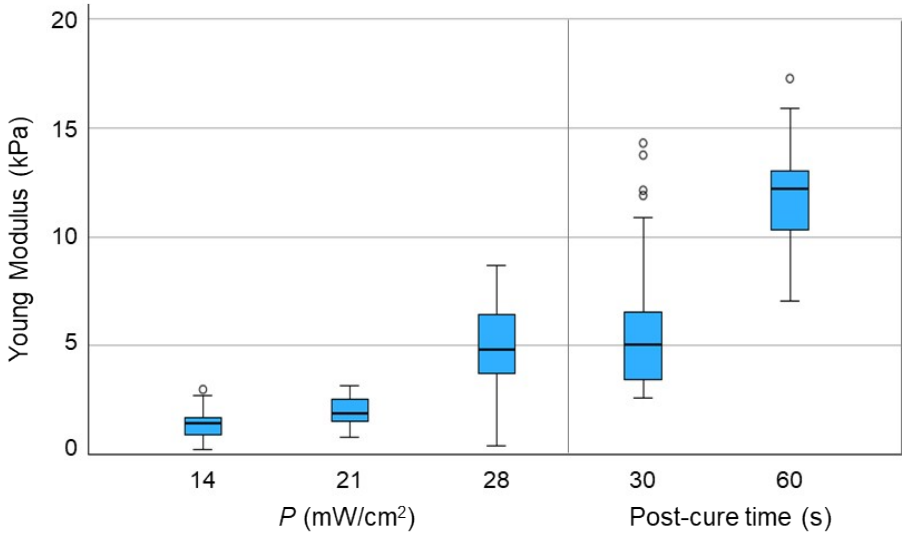

Figure S4. Statistical analysis of AFM measurements of Young's modulus using independent-samples Kruskal-Wallis test.

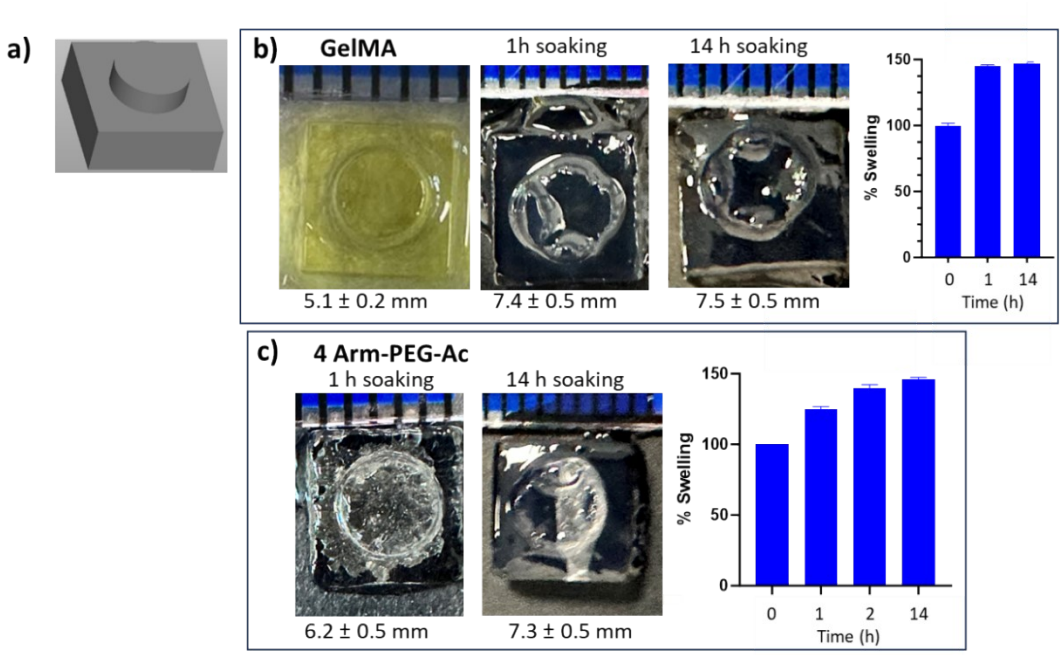

Figure S5. Degree of swelling analysis of hydrogels printed into **(a)** design with sizes of 5 x 5 x 3 mm and photographs of **(b)** printed GelMA and **(c)** 4Arm-PEG-Ac soaked in water at different time points. (Inset in b, c) Degree of swelling of GelMA, and 4Arm-PEG-Ac, respectively.

SI2. Two photon photoplolymerization manufacturing

The processability window for 2PP was assessed by printing an array of 100 µm x 100 µm x 100 µm cubes and lattice structures (cell dimension 20 µm). In the array, the power and printing speed were varied between 25 and 300 mW and 100 and 800 mm/s, respectively, while the layer height and the hatching distance were set at 1 µm.

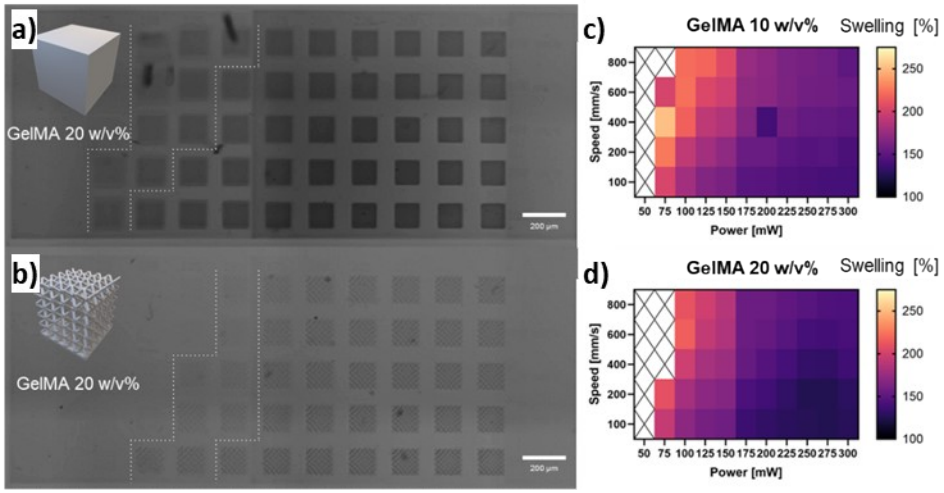

Figure S6. Processability window of GelMA 10 w/v% and GelMA 20 w/v%. **(a, b)** GelMA 20 w/v%. Array cubes **(a)** and lattices **(b)** of 100 x 100 x 100 µm printed at varying power (50-300 mW) and speed (100 - 800 mm/s). Images obtained via simulated bright field confocal imaging. **(c, d)** Top swelling of the cubes of the GelMA 10 w/v% ink **(c)** and GelMA 20 w/v% ink computed as the ratio between the area of the printed samples and the nominal area.

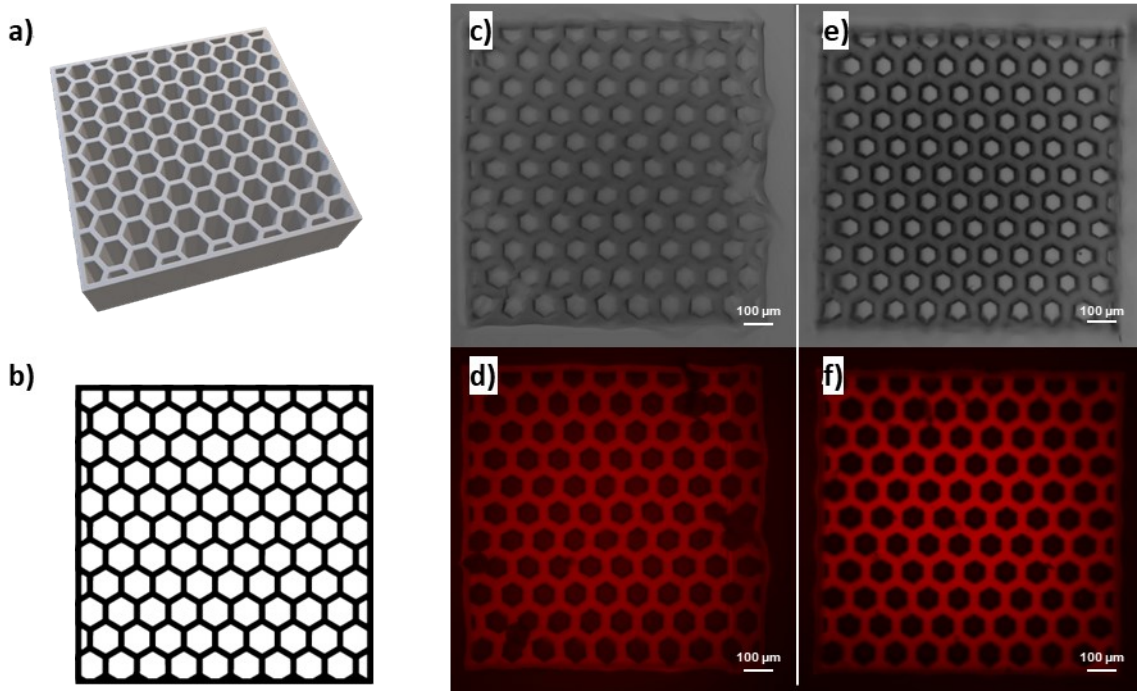

Figure S7. MPL printed structures using GelMA. **a)** STL file of the honeycomb structures (external dimensions: 1000 x 1000 x 300 µm). **b)** Crossection of the stl file corresponding to the following images of the construct. Honeycomb structures printed using GelMA 10 w/v% **(c, d)** and GelMA 20 w/v% **(e, f)** stained using rhodamine B (RhB) 1 mg/mL (z = 100 µm). Simulated bright field images **(c, e)** and RhB signal **(d, f)**.

SI3. Biocompatibility assessment – preliminary studies

Preliminary assessment of biocompatibility of hydrogels was performed using live-dead assay on fibroblast-like cell line L929 exposed for 24h to GelMA (up to 10%), with optical images revealing no noticeable changes in cell density and cell morphology, and the cell viability higher than 70% (Figure S6).

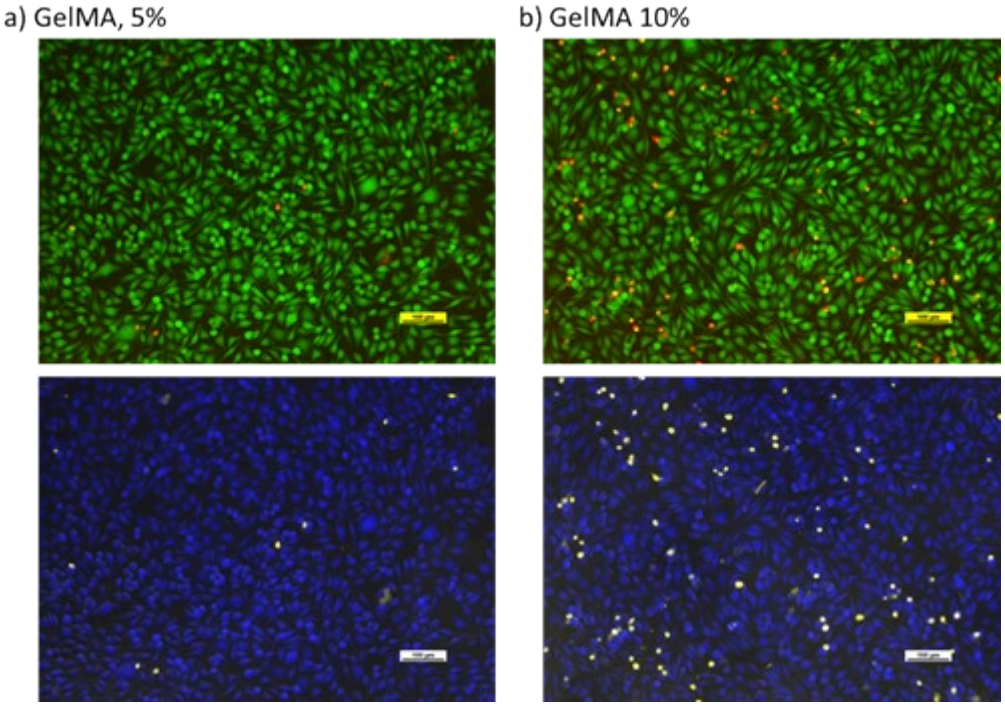

Figure S8. Fluorescent microscopy images of L929 fibroblast cells exposed for 24h to (a) 5% and (b) 10% GelMA.