## **SUPPLEMENTARY INFORMATION**

## **Experimental Procedure**

*Materials*. Poly(ethylene glycol) (PEG, MW = 10000 Da), Poly(ethylene glycol) mono methyl ether (PEGME, MW = 5000 Da) and [2-(Methacryloyloxy)ethyl]dimethyl-(3-sulfopropyl)ammonium hydroxide (DMAPS, 95%) were obtained from Sigma-Aldrich. 2-Butanone (>99%), Ethyl Phenyl(2,4,6-trimethylbenzoyl)phosphinate (>95%), phenylbis(2,4,6-trimethylbenzoyl)phosphine oxide (TPO, >96%), acrylic acid (AA, >99%) and glacial acetic acid (>99.5%) were obtained from TCI America. Diacetone Acrylamide (DAAm, 99%) and N,N'-Methylenebisacrylamide (MBAc, 97%) were obtained from Alfa Aesar. Lithium bromide (>99%), 4-Dimethylaminopyridine (DMAP, >99%), and 3-(3-Dimethylaminopropyl)-1-ethyl-carbodiimide hydrochloride (EDC, >99%) were obtained from Chem Impex International. Acetone (>99.5%) was obtained from Fisher Scientific. 4-cyano-4- (((dodecylthio)carbonothioyl)thio)pentanoic acid (DCT, 97%) was obtained from Boron Molecular. Phenol Red (~95%) was obtained from Aldrich Chemical Co.

**Synthesis of Lithium phenyl-2,4,6-trimethylbenzoylphosphinate (LAP) photoinitiator.** To a 250 mL beaker containing 100 mL of 2-Butanone, Ethyl Phenyl(2,4,6-trimethylbenzoyl)phosphinate (5.73 g, 18.1 mmoles) and lithium bromide (6.30 g, 72.7 mmoles) were added and stirred into solution. Once dissolved, the mixture was heated in a 60°C water bath for 24 hours. The precipitated product was vacuum-filtered, washed four times with 2-butanone, and vacuumed dry to obtain the pure white solid (3.47 g, ~65% yield).

*Synthesis of difunctional PEG macro-CTA.* To a 300 mL beaker containing dichloromethane (~100 mL), PEG (8.26 g, 0.826 mmoles), DCT (1.00 g, 2.48 mmoles), and DMAP (0.302 g, 2.48 mmoles) were added and magnetically stirred into solution. Once homogenized, EDC (0.475 g, 2.48 mmoles) was slowly fed into the solution, and the mixture was allowed to react overnight. Following reaction, the mixture was dialyzed in SpectraPor Regenerated Cellulose Dialysis tubing (6 - 8 kDa cutoff) against acetone for 3 days to remove any excess DCT. This was followed by dialysis against distilled water to remove residual acetone. The purified solution was frozen and lyophilized to obtain the final product.

*Synthesis of monofunctional PEG macro-CTA.* To a 100 mL beaker containing dichloromethane (~50 mL), PEGME (4.13 g, 0.825 mmoles), DCT (0.500 g, 0.124 mmoles), and DMAP (0.151 g, 0.124 mmoles) were added and magnetically stirred into solution. Once homogenized, EDC (0.237, 0.124 mmoles) was slowly fed into the solution, and the mixture was allowed to react overnight. Following reaction, the mixture was dialyzed in SpectraPor Regenerated Cellulose Dialysis tubing (3.5 kDa cutoff) against acetone for 3 days to remove excess DCT. The solution was then dialyzed against water for two days, frozen and lyophilized.

*Characterization of difunctional PEG macro-CTA.* Difunctional PEG macro-CTA was characterized via <sup>1</sup>H Nuclear Magnetic Resonance (NMR, Bruker 400MHz) and UV-Vis Spectrometry (Tecan Infinite M Nano+). <sup>1</sup>H NMR in CDCl<sub>3</sub> was used to identify the appearance of the ester peak (4H, ~4.2 ppm) that forms during reaction between PEG and DCT RAFT agent. Integration of these ester protons with respect to the methylene protons of PEG (~3.8 ppm) indicated the efficiency of the esterification reaction. UV-Vis was used to quantify the CTA density per gram of polymer for the synthesized macro-CTAs (10 mg

mL<sup>-1</sup> in solution) utilizing DCT as a standard curve in 70% isopropanol at a wavelength of 325 nm. GPC of samples were measured using DMF + 0.1 wt% LiBr as mobile phase and PS as calibration standards.

DAAm PISA UV exposure studies with monofunctional PEG macro-CTA. In an effort to show that physical crosslinking of PISA resins is a result of multi-functional RAFT macro-CTAs, UV curing trials of PISA resins that employed monofunctional RAFT macro-CTAs were studied. CTA density of the macro-CTA was determined via UV-Vis using Beer's Law standard curve. The DP (degree of polymerization) of the chain-extended solvophobic block (DAAm) was varied (DP 250, 500, 750, 1000). Total solids concentration for the prepared monofunctional resins was fixed at 40 wt. %, and photoinitiator concentration was kept fixed at 0.5 wt. % with respect to solids. DAAm monomer, monofunctional PEG macro-CTA and distilled water were added to a 50 mL tube and vortexed until homogeneous. LAP photoinitiator was prepared as a stock in distilled water at a concentration of 50 mg mL<sup>-1</sup> and then added to the 50 mL tube containing the resin. 600  $\mu$ L of each resin with varied DP was pipetted in the middle of the 3D DLP printer resin vat and exposed under UV light for 2 minutes on the printer. Following UV exposure, the resin drops were lightly scraped to see if they held their shapes. A representative procedure for preparation of DP 250 DAAm resin is as follows: to a 50 mL tube, 1 g difunctional PEG macro-CTA (0.5 g, 0.063 mmoles DCT), DAAm (2.66 g, 15.8 mmoles), and distilled water (4.74 g) were added and vortexed. 316  $\mu$ L of a 50 mg mL<sup>-1</sup> LAP stock in water was then added to the resin and vortexed. Results of monofunctional macro-CTA studies can be found in SI Figure 2.

**PISA DLP printing of crosslinked and uncrosslinked resins using difunctional PEG macro-CTA.** To synthesize 3D DLP PISA resins, the CTA density per gram polymer (moles DCT / g PEG macro-CTA) was used to target a degree of polymerization for the PISA 3D printing resin. This value was 0.139 mmoles DCT/g polymer (75% theoretical). In terms of resin formulation, hydrophilic difunctional PEG macro-CTA was blocked either with DAAm under aqueous conditions or with acrylic acid and DMAPs in acetic acid. DAAm monomer is soluble in water but its homopolymer is not. The target degree of polymerization for this PISA was 500 as a total solids concentration of 40 wt. %. The target degree of polymerization (DP) of 500 is based on the total number of CTAs present in the polymer (DP 500 from both arms of the RAFT agent). The CTA graft density was determined via UV-Vis .LAP was employed as a water-soluble photoinitiator at a concentration of 0.25 wt. % with respect to solids. Phenol red was additionally employed as a photoabsorber at a concentration of 0.01 wt. % with respect to solids. Two resins were prepared and 3D-printed under PISA conditions: one with and without inclusion of MBAc crosslinker. In the case of the MBAc-containing crosslinker, MBAc was included at a concentration of 2.5 wt% with respect to solids.

Similarly, PISA resins were formulated by blocking AA and DMAPS to difunctional PEG macro-CTA in acetic acid. DMAPS monomer is soluble in acetic acid but its polymer is not. However, DMAPS alone was slow to block and formed a very sticky print that would get stuck to the resin vat. Addition of AA seemed to speed up curing and reduce stickiness of the cured parts while enough DMAPS was still present in the formulation to promote self-assembly. In this case, target degree of polymerization for the AA was 550 and for the DMAPs was 300 all at a total solids concentration of 35 wt. %. TPO was employed as the photoinitiator in this case at a concentration of 0.75 wt. % with respect to solids. Phenol red, sparingly soluble in acetic acid, was still employed as the photoabsorber at a concentration of 0.01 wt. % with respect to solids, though the phenol red was more suspended than dissolved. Much like with the DAAm formulation, two resins were prepared: one with and without MBAc crosslinker in a manner similar to what was described above.

Resins were filtered through a fine mesh into the resin vat. ChituBox slicing software was used to set printing parameters and prepare the file for printing on the AnyCubic Photon Mono DLP printer. DAAm PISA printing parameters included 100 µm layer thickness, a bottom exposure time of 45 s per layer and a normal exposure time of 40 s per layer. Acrylic acid-co-DMAPS PISA printing parameters included 100 µm layer thickness, a bottom exposure time of 50 s per layer and a normal exposure time of 45 s per layer. Prints included tree frogs for DAAm and acrylic acid-co-DMAPS resins in addition to an ASTM D638 Type V dogbone for DAAm resins. A representative procedure for DAAm PISA resin preparation without crosslinker is as follows: to a 50 mL conical tube, difunctional PEG macro-CTA (1 g, 0.139 mmoles DCT), DAAm (11.7 g, 69.3 mmoles), LAP (31.8 mg, 0.108 mmoles), and phenol red (1.27 mg, 3.59 µmoles) were dissolved homogeneously in distilled water (19.1 g). A representative procedure for acrylic acid-co-DMAPS PISA resin preparation without crosslinker is as follows: to a 50 mL conical tube, difunctional PEG macro-CTA (1 g, 0.139 mmoles DCT), acrylic acid (5.50 g, 76.3 mmoles), DMAPS (13.44 g DMAPS, 48.2 mmoles), and TPO (150 mg, 0.357 mmoles) were dissolved in acetic acid (37.0 g). Phenol red (1.99 mg, 5.63 µmoles) was additionally added in but stayed suspended in acetic acid.

**Dissolution studies of 3D PISA prints.** Following 3D printing of PISA resins with and without crosslinker, PISA printed tree frogs using DAAm were placed into DMF for dissolution studies. DMF was chosen since it should solubilize both PEG and DAAm blocks of the PISA polymer. The printed tree frog without crosslinker and printed tree frog containing MBAc crosslinker were placed in separate 100 mL beakers containing 50 mL of DMF. Dissolution (or lack thereof) was documented for 4 hours. Additionally, PISA printed tree frogs using acrylic acid and DMAPS in acetic acid were placed into 0.5 M NaCl aqueous solution for dissolution studies. The salt solution was chosen as it should solubilize PEG, acrylic acid and DMAPS. Frogs were placed into 50 mL salt solution, and dissolution was followed for two to four hours.

*Characterization of 3D printed PISA polymers.* PISA polymers were characterized via <sup>1</sup>H Nuclear Magnetic Resonance (NMR), Scanning Electron Microscopy (SEM), Gel Permeation Chromatography (GPC) and Atomic Force Microscopy (AFM). <sup>1</sup>H NMR of the starting resins of DAAm and AA-co-DMAPS that contained no MBAc crosslinker were measured in D<sub>2</sub>O. After dissolution of the frogs for both DAAm PISA (in DMF) and AA-co-DMAPS PISA (in aqueous brine solution), <sup>1</sup>H NMR of the dissolved solutions of DAAm frogs and acrylic acid-co-DMAPs frogs were taken in  $CDCl_3$  and  $D_2O$ , respectively. In doing so, conversion of the monomer in the post-printed part could be determined by measuring the disappearance of the vinyl resonances (5.5 - 6.5 ppm depending on the monomer) with respect to the methylene protons in the PEG backbone (OCH<sub>2</sub>CH<sub>2</sub>, ~3.6 ppm). Additionally, <sup>1</sup>H NMR of lyophilized, uncrosslinked PISA polymers for both DAAm and AA-co-DMAPS were measured in CDCl<sub>3</sub> and D<sub>2</sub>O, respectively. <sup>1</sup>H NMR of the pure materials helped to estimate composition and degree of polymerization for the blocked polymer, which was later confirmed by GPC. SEM imaging of the lyophilized DAAm PISA 3D printed part was performed by first sputter coating the fractured surface with Au using Denton sputter coater. Microstructure of the coated surface was then imaged on the PRISMA E SEM at a voltage of 20 kV. GPC of DAAm dissolved frogs samples were measured using DMF + 0.1 wt% LiBr as mobile phase and PS as calibration standards. GPC of AA-co-DMAPS dissolved frogs was not measured due to lack of a suitable mobile phase for both PISA blocks. GPC results of the post-printed dissolved DAAm uncrosslinked frog are shown in SI Figure 2. Atomic Force Microscopy (AFM) was performed on a Dimension Icon system (Bruker) using the Scanasyst air mode. To prepare the samples, 10uL of the freshly prepared polymer resin was deposited onto a freshly cleaved mica surface and cured under UVlight for 1 minute, then drying for another 30 minutes. The Scanasyst mode with ascan rate of

0.8 Hz was used to determine the surface profiles of the resins with ultrasharp 14 series (NSC 14) tips purchased from Nanoandmore.

*Mechanical properties of 3D-printed PISA dogbones.* DAAm PISA resins with and without crosslinker were cast into a plastic ASTM D638 Type V dogbone mold and cured for 2 minutes under UV light. After demolding, the part were immediately loaded onto the Shimadzu EZ-LX series tensile tester using 5kN load cell tensile grips. Stroke was set to a value of 10 mm min<sup>-1</sup>, and parts were placed under tension until failure. Modulus of elasticity for the materials was determined by taking the slope of the linear portion of the stress-strain curve that intersects the origin. % elongation was determined using the displacement of the dogbone gauge length relative to its initial gauge length. Results of mechanical properties are summarized in SI Table I and SI Figures 7 and 8. AA-co-DMAPS PISA resins with and without crosslinker were also cast into dogbones, dialyzed against isopropanol to remove the acetic acid and measured. While the results were reported, the large inconsistencies that resulted for the uncrosslinked parts were attributed to the rapid drying of the isopropanol in the part as it was being measured. Due to the rapid drying, the part became stiffer during measurement. It should be noted that these results, although they show a general trend, may be unreliable.

**Controlled dissolution studies of 3D printed PISA P scaffolds.** DAAm PISA resins with and without crosslinker were prepared as described previously. Two separate parts incorporating DAAm resins with and without crosslinker were printed to test controlled dissolution of the part architecture. CAD files for these two designs are shown in SI Figure 7. In the case of the platform and pillar scaffold design (SI Figure 7A), the DAAm PISA resin containing crosslinker was used to print the platforms while the resin containing no crosslinker was used to print the pillars. In the case of the platform-cylinder design (SI Figure 7B), the resin containing crosslinker was used to print the platform while the one without crosslinker was used to print the cylinders. ChituBox slicing software was used to determine the slices at which the design transitioned from a platform to pillar, which indicated the slice at which the resin needed to be swapped from the resin containing crosslinker to the one without crosslinker. This was repeated multiple times by stopping the print at the appropriate layer, cleaning the vat to avoid contamination and switching out the resin before finally resuming the print. The same process was repeated with the platform-cylinder design. Following printing, the parts were washed with distilled water and fully submerged in DMF for dissolution. Dissolution of the pillars and cylinder were closely followed while the platforms remained intact.

## **SI Results**

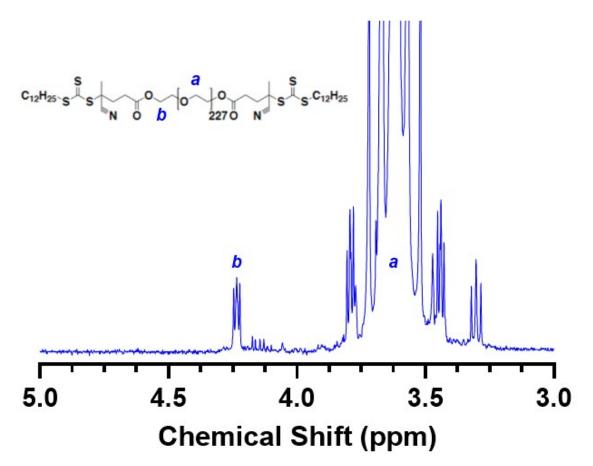
In addition to DAAm PISA P system described in the main manuscript, we also evaluated PISA P from the difunctional PEG macro-CTA in acetic acid using a 70:30 (by weight) mixture of DMAPS and acrylic acid (SI Figure 3A). This system was selected as we have previously shown that acetic acid solubilizes both hydrophilic monomers like DMAPS and hydrophobic monomers such as lauryl methacrylate, which then phase separate when polymerized from an appropriate macro-CTA to form stable nanostructures.<sup>1</sup> Acrylic acid was added to DMAPS formulations in order to reduce the tackiness of the parts during the printing process. An initial monomer concentration of 33 wt. % was used targeting a theoretical DP of 850 (70 wt. % DMAPS, 30 wt. % AA). The TPO and phenol red concentrations for these resins were 0.75 wt. % and 0.01 wt. %, respectively. Printing parameters for DMAPS / AA system were as follows: cure layer thicknesses of 100 µm, bottom exposure times of 50 s and normal exposure times of 45 s.

3D printing studies were conducted either without crosslinker or with 2.5 wt. % MBAc. In these experiments, both formulations yielded successful prints with no noticeable defects (SI Figures 3B and 3C). While both DMAPS and acrylic acid are soluble in acetic acid, the resultant copolymers form gels upon polymerization. Because of the low solubility of the sulfobetaines in deionized water, the resultant poly(AA-co-DMAPS) copolymers were placed in an aqueous salt solution consisting of 1 M sodium bicarbonate and 0.5 M NaCl. Sodium bicarbonate was added in order to neutralize the acrylic acid residues while the high salt solution was added to screen interactions between the zwitterionic DMAPS residues.<sup>2,3</sup> The frog that contained no crosslinker fully dissolved within four hours (SI Figure 3B). The frog containing 2.5 wt. % crosslinker did not dissolve but did swell up to twice its original weight in water (SI Figure 3C).

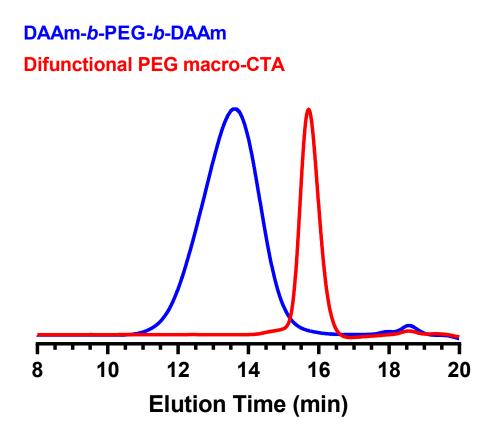
Mechanical properties for the AA-co-DMAPS PISA resins were also measured. ASTM D638 Type V dogbones were mold-cured (like the DAAm system) for this system. Due to the presence of acetic acid in the resin, the dogbones were dialyzed against isopropanol following mold curing to make them safer to handle during testing. Results for AA-co-DMAPS dogbones with and without crosslinker are shown in SI Table I and SI Figure 8. While the results displayed a similar trend as DAAm for crosslinked (higher modulus, lower elongation) v. uncrosslinked (lower modulus, higher elongation) dogbones, the results overall showed inconsistencies, especially for the uncrosslinked dogbones. This was attributed to quick drying of the part during testing due to parts being solvated with isopropanol. There was also high variability in the values obtained for elongation of the uncrosslinked dogbones. The quick drying also led to the parts being stiffer during testing, which could explain why the modulus for the crosslinked of this system was much higher compared to that of the one for the DAAm system. For these reasons, the DAAm system property values should not be directly compared to the AA-co-DMAPS property values.

It should be noted that while phenol red was included in these studies as a photoabsorber<sup>4</sup>, it has been shown by Bagheri *et. al.* that high resolution can also be achieved in RAFT-based 3D printing due to the light absorption provided by the RAFT agent itself.<sup>5</sup> We explored this by printing hexagonal lattices with and without the use of photoabsorber. This is shown in SI Figure 9. The hexagonal lattices printed without the use of phenol red overall had mostly high resolution (well-defined hexagons) though there was evidence of overcuring into the hollow hexagonal centers. The one printed with phenol red showed well-defined hexagons with no overcuring into the hexagonal centers. While there is a slight improvement with the employment of the phenol red (no overcuring in the hexagons), it was shown that RAFT-based 3D printing resins can produce similarly high resolution, indicating phenol red may not be necessary.

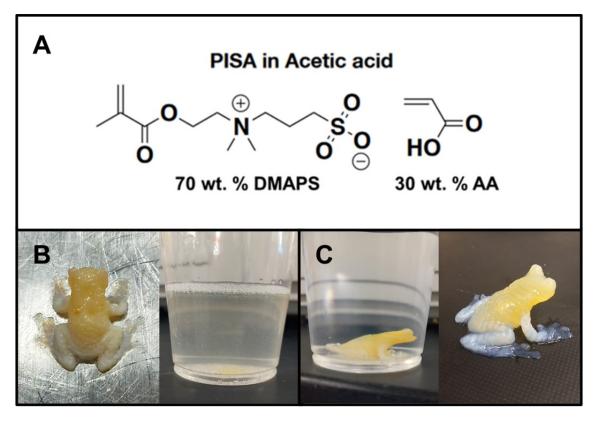
Finally, <sup>1</sup>H NMR of the purified dissolved frog polymers were measured and are shown in SI Figure 10. These measurements were conducted in CDCl<sub>3</sub> and D<sub>2</sub>O for DAAm and AA-co-DMAPS system, respectively. For the DAAm system, DP was estimated by integrating the methyl protons (6H, ~1.3 ppm) on the carbon atom adjacent to the amide bond of the diacetone acrylamide block with respect to the PEG backbone methylene protons (~3.6 ppm). For AA-co-DMAPS system, DP was estimated by integrating the methyl protons adjacent to the quaternary ammonium (6H, ~3.1 ppm) on poly(DMAPS-co-AA) with respect to PEG methylene protons (~3.6 ppm). GPC molecular weight values were measured for the DAAm system (but not AA-co-DMAPS system due to lack of a suitable solvent). The values obtained from GPC were compared against the NMR data to see how accurate the estimated value was.



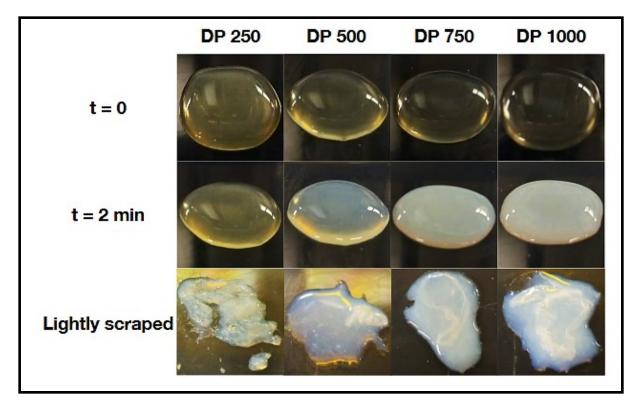
SI Figure 1. <sup>1</sup>H NMR of synthesized difunctional PEG macro-CTA. Resonance (a) corresponds to the methylene protons of the PEG backbone repeat unit (OCH<sub>2</sub>CH<sub>2</sub>, 3.6 ppm) whereas (b) corresponds to the methylene protons adjacent to the newly formed ester peak. This ester peak is the result of the reaction of the DCT RAFT agent acid group to the hydroxyl-terminated ends of the PEG. Integrations suggest a reaction efficiency of ~77%. This matches up closely with the CTA value obtained from UV-Vis which was determined to be 75% of theoretical. This indicates the presence of some monofunctional PEG in addition to difunctional PEG.



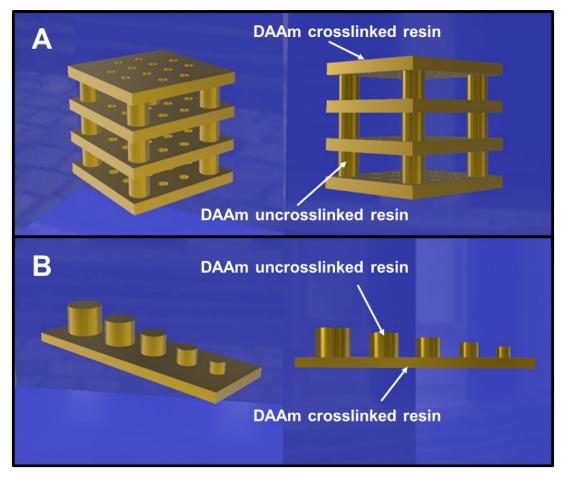
**SI Figure 2.** GPC traces for difunctional PEG macro CTA (red) and DAAm PISA dissolve frog polymer (blue) polymerized from this difunctional RAFT PEG macro CTA. The measured molecular weight (Mn) was determined to be 177886 Da, close to the targeted value of 179000 Da. The wide PDI is believed to possibly be attributed to the presence of a high radical flux when compared to conventional thermal RAFT polymerizations. It is also believed the high intensity of the UV light, which can degrade CTAs, could have contributed to the wide distribution.



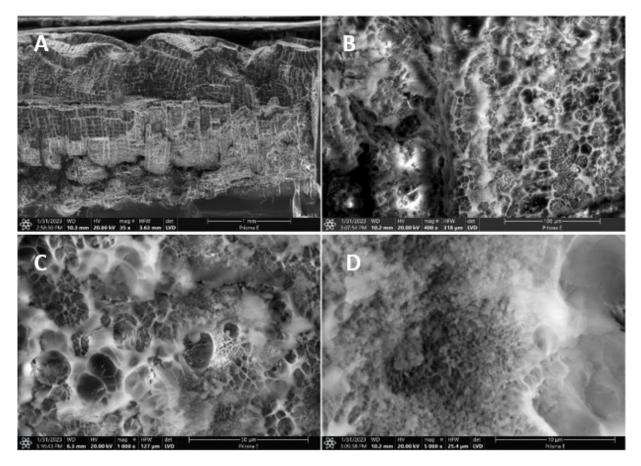
SI Figure 3. Results for PISA printing of AA and DMAPS from difunctional PEG macro-CTA in acetic acid. (A) Scheme for PISA 3D printing with a resin formulation that utilized DMAPS and AA monomers. (B) AA-co-DMAPS PISA 3D printed frog without crosslinker, insoluble in acetic acid, dissolved in aqueous brine solution. This is an indication that physical crosslinking is holding the part together. (C) AA-co-DMAPS PISA 3D printed frog with crosslinker did not dissolve in the aqueous brine solution due to the presence of chemical crosslinks, though it did swell to approximately twice its own weight.



**SI Figure 4**. Results for monofunctional PEG macro-CTA PISA studies with DAAm in water. As shown, all resins successfully underwent PISA when exposed to UV light as evidenced by the increase in opacity with increasing DP. Resins were lightly scraped following 2-minute UV exposure. Although the DP 250 resin slightly gelled, it failed to hold its shape following exposure which indicates little if any physical crosslinking. DP 500, 750 and 1000 resins showed little if any evidence of gelation and were very fluid. These studies show the use of multi-functional RAFT agents may be needed to promote the physical crosslinking necessary for the exposed resin to cure and hold its shape during 3D DLP printing.



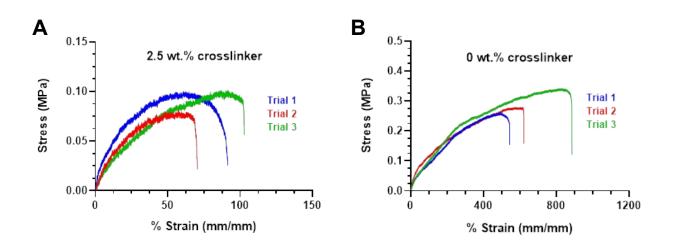
**SI Figure 5.** CAD files for 3D scaffolds used for controlled dissolution tests. (A) shows the CAD file for 3D printable platform stack in which the uncrosslinked DAAm resin was used to print the pillars and the crosslinked DAAm resin was used to print the pillars dissolved away while the platforms swelled but remained intact. (B) shows the CAD file for cylindrical structures on a rectangular platform. In this case, the cylinders were printed using the DAAm resin containing no crosslinker while the platform itself was made using the crosslinked DAAm resin. Upon printing the scaffold and placing in DMF, the cylinders dissolved away but the platform remained intact. Results for the printed objects are shown in Figure 2.



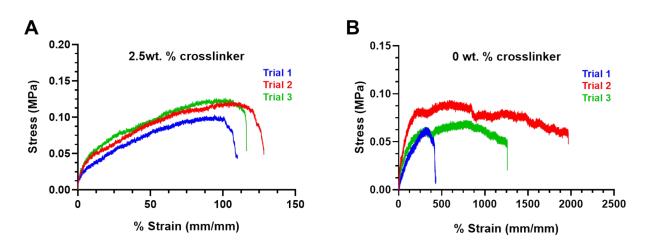
**SI Figure 6**. SEM images of purified, dissolved DAAm PISA frog that contained no crosslinker. As shown, the microstructure consists of a series of web-like architectures with high porosity.

PISA resin composition	E (kPa)	Elongation at break (%)
DAAm (0 wt. % crosslinker)	390 ± 61	682 ± 178
DAAm (2.5 wt. % crosslinker)	763 ± 20	87 ± 17
AA-co-DMAPS (0 wt. % crosslinker)	1527 ± 45	114 ± 11
AA-co-DMAPS (2.5 wt. % crosslinker)	231 ± 65	1300 ± 964

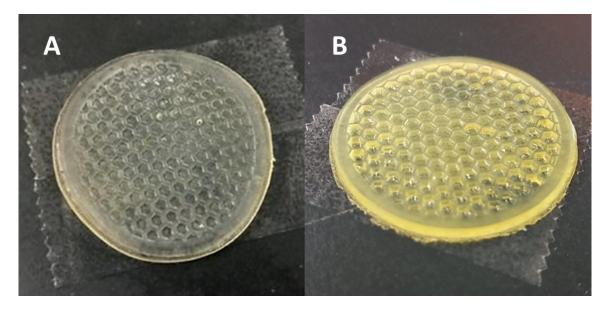
SI Table I. Mechanical properties for DAAm resins with and without crosslinker



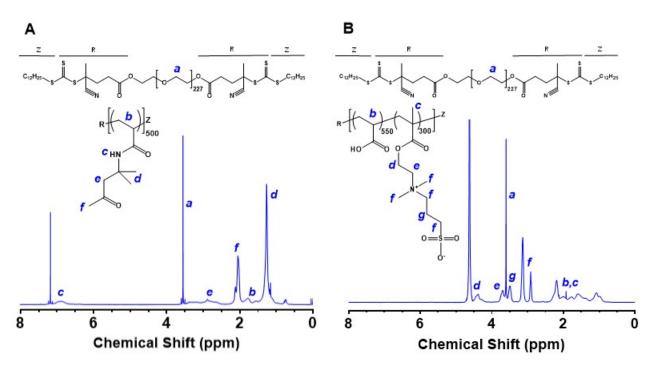
SI Figure 7. Mechanical properties for DAAm PISA dogbones. (A) Results for dogbones cured from DAAm PISA resins containing 2.5 wt. % MBAc crosslinker (chemically-crosslinked) . (B) Results for dogbones cured from DAAm PISA resin containing no MBAc crosslinker (physically-crosslinked).



SI Figure 8. Mechanical properties for AA-co-DMAPS PISA dogbones. (A) Results for dogbones cured from AA-co-DMAPS resins containing 2.5 wt. % MBAc crosslinker (chemically-crosslinked). (B) Results for dogbones cured from AA-co-DMAPS PISA resin containing no MBAc crosslinker (physically-crosslinked).



**SI Figure 9.** Effect of phenol red on printing resolution of DAAm PISA resins. (A) DAAm PISA part printed without the use of photoabsorber. While it achieved high resolution, the resin overcured into the hexagons. (B) DAAm PISA part printed with the use of phenol red photoabsorber showed high resolution and no overcuring of the resin into the hexagonal crevices occurred.



**SI Figure 10.** <sup>1</sup>H NMR spectra for purified triblock PISA polymers for (A) DAAm and (B) AA-co-DMAPS blocked from difunctional PEG macro CTA.

## **SI References**

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