Programmable Anisotropy and Percolation in Supramolecular Patchy Particle Gels

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ABSTRACT: Patchy particle interactions are predicted to facilitate the controlled self-assembly and arrest of particles into phase-stable and morphologically tunable “equilibrium” gels, which avoids the arrested phase separation and subsequent aging that is typically observed in traditional particle gels with isotropic interactions. Despite these promising traits of patchy particle interactions, such tunable equilibrium gels have yet to be realized in the laboratory due to experimental limitations associated with synthesizing patchy particles in high yield. Here, we introduce a supramolecular metal-coordination platform consisting of metallic nanoparticles linked by telechelic polymer chains, which validates the predictions associated with patchy particle interactions and facilitates the design of equilibrium particle hydrogels through limited valency interactions. We demonstrate that the interaction valency and self-assembly of the particles can be effectively controlled by adjusting the relative concentration of polymeric linkers to nanoparticles, which enables the gelation of patchy particle hydrogels with programmable local anisotropy, morphology, and low mechanical percolation thresholds. Moreover, by crowding the local environment around the patchy particles with competing interactions, we introduce an independent method to control the self-assembly of the nanoparticles, thereby enabling the design of highly anisotropic particle hydrogels with substantially reduced percolation thresholds. We thus establish a canonical platform that facilitates multifaceted control of the self-assembly of the patchy nanoparticles en route to the design of patchy particle gels with tunable valencies, morphologies, and percolation thresholds. These advances lay important foundations for further fundamental studies of patchy particle systems and for designing tunable gel materials that address a wide range of engineering applications.

KEYWORDS: patchy particles, self-assembly, supramolecular chemistry, equilibrium gels, bioinspired materials

INTRODUCTION

The route to dynamic arrest in particle gels has profound consequences on their emergent structure and mechanical properties, and has important implications for understanding self-assembly in biology and optimizing the design of functional soft materials. For particles with isotropic and reversible interactions such as colloids interacting via depletion interactions, dynamic arrest (at volume fractions \( \phi \) below the glass transition) commonly occurs through an arrested spinodal decomposition, following quenching into a two-phase coexistence region.1−7 In contrast, for particles with limited-valency interactions, theoretical studies have predicted that arrest can occur through equilibrium associations, due to the emergence of interaction anisotropy and suppression of the two-phase region to higher interaction strength \( \epsilon \) and lower \( \phi \) (Figure 1A).3−7 Unlike isotropic particle gels, which continuously age with time due to their underlying thermodynamic instability, these “patchy” particle gels can thus undergo “equilibrium arrest” and remain thermodynamically stable and thus avoid temporal aging effects. Patchy particle arrest therefore represents a promising method for designing soft particle gels for materials applications requiring long-term stability, with relevance in structural and biomedical engineering. Moreover, theoretical studies have predicted that the morphology and the density of such patchy particle gels should also be tunable by controlling the valency (and therefore the anisotropy) of particle interactions,2,6 enabling the design of stable soft materials with a broad range of microstructures. In fact,
growing evidence suggests that this exact strategy is utilized during the assembly of soft and stable biological materials with tunable microstructures, such as graded-index protein lenses in squid eyes and degradation-resistant egg whites in duck eggs, among others.20,21

Despite these promising traits of patchy particle gels, experimental validation of their properties remains limited. Although advances in molecular engineering now permit the design of a myriad of patchy particles with precise interaction sites and valencies, these systems remain difficult to synthesize and assemble at the requisite scales for designing patchy particle gels. Thus, patchy particle arrest has so far only been studied in a narrow range of experimental systems with fixed valencies, most prominently in discotic clay particles but also in molecular systems, such as DNA stars and peptides.22 Notably, the lack of an accessible experimental particle platform in which the particle patch valency can be tuned has prevented access to the design of gels with the wide ranges of morphologies and percolation behaviors predicted by theory.23,24 A patchy-particle-based material system with tunable valency would not only facilitate bioinspired designs of stable particle gels with tunable morphologies and percolation thresholds but also allow deeper studies of the fundamental physics of patchy particle self-assembly in a variety of environments.2 Thus, in this work, we show that a polymer–metal nanoparticle composite gel system governed by supramolecular metal-coordination interactions provides an experimental realization of a patchy particle system with tunable effective interaction valency, and permits study of the self-assembly and equilibrium arrest of particles at a variety of morphologies and percolation thresholds.

RESULTS AND DISCUSSION

Our gel system consists of 1-arm catechol-functionalized poly(ethylene glycol) (1cPEG) chains, which are grafted onto Fe3O4 nanoparticles (NPs), and 4-arm nitro catechol-functionalized poly(ethylene glycol) (4nPEG) telechelic linker chains (Figure 1B). The catechol ligand coordinates strongly onto the Fe3O4 NP surface to facilitate a highly water-stable 1cPEG-NP complex (Figure 1C), but the ligand can be dynamically exchanged by the stronger binding nitrocatechol ligand on the 4nPEG (Figure 1D,E); analogous control systems with 4-arm PEGs functionalized with weaker affinity ligands that cannot displace the 1cPEG graft on the NPs as effectively are not able to create stiff gels (Figure S5). Step strain measurements of the SPPH at 25 °C reveal slow and broadly distributed stress relaxation modes, which can be succinctly captured by a Kohlrausch–Williams–Watts (KWW) model of the form $G(t) = G_\infty \exp[-(t/\tau)^\alpha]$, with a stretching exponent of $\alpha \approx 0.31$ (parameters in Table S3), consistent with the theoretically predicted $\alpha \approx 0.33$ for a deeply arrested equilibrium associating system (Figure 1G).34 Importantly, stress relaxation measurements after 24 and 48 h of aging nearly overlap with each other, again in support of the negligible rheological aging in our patchy gel, in contrast to the substantial rheological aging observed in typical colloidal gels.34 Characterization of the time-dependent structure factor $S(q)$ in the SPPH via small-angle X-ray scattering (SAXS) tells a similar story (Figure 1H). $S(q)$ is obtained from scattering measurements by dividing the scattered intensity $I(q)$ by a spherical form factor (Figure S7; see also Methods). A pronounced interparticle peak is observed at $q \approx 0.05 \AA^{-1}$ (arrow in Figure 1H), corresponding to an interparticle distance of $D_{\text{np-np}} = 2\pi/q = 12.5 \text{ nm}$, which represents the combined diameters of the particle ($\sim 7 \text{ nm}$, Figure S2B) and the bridging polymer ($\sim 5.5 \text{ nm}$, Figure S1B) and is in good agreement with the proposed microstructure of the SPPH (Figure 1E). The upturn in $S(q)$ at low $q$ indicates the formation of local aggregates with fractal dimensions of $D_f = 2.7$ (as determined by the slope), consistent with commonly observed values in polymer gels and biological patchy gels.8 Characterization of $S(q)$ at even lower $q$ via ultrasmall-angle X-ray scattering (USAXS) reveals that this fractal regime extends up to the local mesh size of $\xi \sim 200 \text{ nm}$ (Figure S6), suggesting that $D_f = 2.7$ originates from intramesh surface scattering of NP aggregates.8 Importantly, unlike arrested spinodal decomposition gels which exhibit continuous structural aging with time,34,54 our SPPHs exhibit no evidence of structural aging as
manifested by the overlap in the $S(q)$ measurements after 24 and 48 h of aging, in agreement with mechanical measurements (Figure 1F,G) and providing strong evidence of equilibrium arrest in the SPPH.

Another distinct hallmark of arrested patchy particle systems is the controllability of their self-assembly and phase behavior a priori by the external tuning of the particle patch valency (Figure 2A).\textsuperscript{2,5,8} We demonstrate such control in our SPPH, wherein the average coordination number $M$ is controlled indirectly by tuning the number of 4nPEG linkers between the particles through the parameter $r = N_{4nPEG}/N_{NP}$. Through electron microscopy and rheology, we show that, at low $r$, the system remains in the sol state as NP self-assembly is anisotropic and sparse (due to small number of linkers); above a percolation threshold of $r$, the system undergoes gelation and can become progressively denser and stiffer with increasing $r$.
Figure 2. Systematic control of self-assembly, morphology, and rheological state behavior of the SPPH obtained by varying the linker-to-NP ratio parameter $r$. (A) Illustration of the effect of varying $N_{\text{ linker}}$ via the linker-to-NP ratio $r$ and therefore the interaction anisotropy by introducing different amounts of 4nPEG linkers to the 1cPEG-NP complex at constant $\phi_{NP}$ and volume (same graphical representations as Figure 1E). (B) Representative SEM micrographs of the SPPH in the gel state at $\phi_{NP} =$ 1.93% and at $r =$ 7.5 and $r =$ 56.0. Scale bar = 100 nm. (C) Representative TEM micrographs of diluted SPPH solutions at $r =$ 3.0 and $r =$ 56.0. The thick strips in the micrographs are the holey carbon grids on which the samples were dried (see Methods). Scale bar = 100 nm. (D) Storage modulus $G''(\omega)$ of the SPPH at $\phi_{NP} =$ 1.93% and various $r$ (corresponding rheograms of the loss modulus $G''(\omega)$ shown in Figure S10A). The viscoelastic gel state at $4.5 < r \leq 56$ shows an almost frequency-independent response in $G''(\omega)$ and $G''(\omega)$, consistent with the broad stress relaxation spectra observed in Figure 1G. (E) Frequency-averaged phase angle $\delta(\omega)$ of the SPPH as a function of $r$ (symbols corresponding to those in panel D). As hypothesized in panel A, increasing $r$ causes the SPPH system to transition from a viscoelastic liquid (shaded in red) to a viscoelastic solid gel (shaded in blue). The onset of phase separation is observed at $r =$ 112. (F) Macroscopic images of the system at $r =$ 168, exhibiting visible phase separation into viscoelastic solid and viscoelastic liquid states. Scale bar = 1 cm.

Increasing $r$; at higher $r$, the dense gel undergoes precipitation due to the increasing isotropy of the NP self-assembly and the expected re-emergence of the two-phase coexistence curve at high valencies (and weaker repulsive interactions) (Figure 2A).^{2,5}

We demonstrate the controllability of the self-assembled microstructure of the SPPH through $r$ via scanning and transmission electron microscopy (SEM/TEM). SEM microcopy images of the SPPH gels (preparation details in the Methods) show dramatic differences at $r =$ 7.5, 28.0, and 56.0, with a notable increase in the extent of dark spots at lower $r$ values (Figures 2B and S9A,B,E). These dark spots represent electron-density-deficient (and thus NP-deficient) regions, which is corroborated by backscattered electron micrographs of the same systems (Figure S9C,D). Thus, the SEM images provide corroborative evidence of the expected microstructures of the SPPH as a function of $r$ (Figure 2A). TEM images of the NP assemblies reveal the microscopic origins of these gel morphologies observed in SEM. For TEM imaging, the SPPHs were imaged in the solution state via significant dilution followed by air-drying on a carbon grid (see Methods). Notwithstanding the effects of the air-drying procedure on the microstructure of the system, a clear difference between the $r =$ 3.0 and the $r =$ 56.0 can be seen (Figures 2C and S8A,B). A sheet-like single layer of NPs is observed for $r =$ 3.0, whereas NP aggregation is observed for $r =$ 56.0, evidenced by the sample opacity due to lack of beam penetration. Similar NP aggregations are observed at even higher valencies facilitated by $r =$ 112 (Figure S8C). We note that sheet-like layers are an important anisotropic structural motif in the SPPH—although they are not visible in the SEM micrographs discussed thus far (Figure 2B), we revisit this issue later and show that these sheet-like features do become salient and visible in SEM upon introducing a network-stabilizing competing ion. Overall, the TEM images provide microscopic evidence of the onset of anisotropy at lower $r$ and vice versa, corroborating the different microstructures of the gels observed via SEM. These demonstrations of programmable morphologies in the SPPH is consistent with predicted behavior of patchy particle systems, where the interaction valency dictates the anisotropy of interactions and thus the density of the resulting self-assembled particle network.^{2,5,5a,5b,9}

Rheological characterizations show that the SPPH system is able to access a wide array of rheological states, as a direct consequence of the different microstructural self-assembly of the NPs at different $r$. Linear viscoelastic measurements show that with increasing $r$ at constant $\phi_{NP} =$ 1.93%, that is, with the same density of cross-linking nodes (the nanoparticles), but with varying density of linkers, the SPPH transitions from a viscoelastic fluid ($r =$ 4.5) to a viscoelastic gel ($4.5 < r \leq 56$) and then transitions back to a softer gel at higher interaction valencies ($r =$ 112) (Figure 2D). This behavior is concisely illustrated by the frequency-averaged phase angle $\delta(\omega)$ of each system: with increasing $r$, the SPPH transitions from a viscous liquid ($\delta(\omega) \approx 90^\circ$) to an elastic solid ($\delta(\omega) \approx 0^\circ$) and back toward the viscous liquid state (Figure 2E). The stiffening of the SPPH at $\phi_{NP} =$ 1.93% with increasing $r$ is consistent with the expected behavior of a patchy system with varying
interaction valencies; for example, the phantom network theory shows that for a valency-controlled network with the same node density, the modulus scales with \((M - 2)/M\). Re-entrant softening is observed at \(r > 56\), which is indicative of an onset of phase separation due to the high interaction valency in the network; this is visibly manifested at \(r = 168\), at which point the SPPH is no longer homogeneous and visibly phase separates into viscoelastic sol and gel phases (Figure 2F; rheological characterization of constituents in Figure S10B). This provides further proof that the re-entrant softening of the SPPH at \(r = 112\) originates from phase separation, as expected from the emergence of the two-phase coexistence at high interaction valency.\(^5\) The \(r\)-dependent state behaviors of the SPPH is therefore consistent with systems composed of patchy particles at different valency (Figure 1A) and suggest a direct relationship between the linker-to-NP ratio, the tunable local anisotropy, the programmable morphology of the self-assembling patchy particle network, and ultimately the final global state of the SPPH system.\(^3,5,6,8,9\)

The valency-dependent rheological state behaviors of our SPPH system can be systematically explored through gelation experiments at different polymer linker concentrations \(n_l\) and NP concentrations \(\phi_{\text{NP}}\), the results of which are summarized by a state diagram of \(r\) as a function of \(\phi_{\text{NP}}\) (Figure 3A). The state diagram shows that high linker-to-NP ratios \(r (N_{\text{4armPEG}} \gg N_{\text{NP}})\) lead to the precipitation and collapse of the gel phase across a wide range of \(\phi_{\text{NP}}\) which is consistent with the predicted emergence of the two-phase coexistence (Figure 1A) and the densification of morphologies in the SPPH (Figure 2A–C) at high interaction valencies. Therefore, at a given linker concentration \(n_l\) the percolation threshold for the gel phase as a function of \(\phi_{\text{NP}}\) is dictated by the point at which \(r = 10.81n_l/\phi_{\text{NP}}\) is suppressed below the threshold for precipitation (roughly when the iso-\(n_l\) lines cross the \(r \approx 100\) precipitate–gel boundary with increasing \(\phi_{\text{NP}}\), Figure 3A). This observation suggests that the NP percolation threshold may be tunable by adjusting the interaction valency \(n_l\) of the linker concentration \(n_l\) and indeed, rheology measurements of the SPPH across different \(n_l\) and \(\phi_{\text{NP}}\) validate this hypothesis (Figures 3B and S13). The frequency-averaged storage modulus \(G'(\omega)\) is obtained by averaging \(G'(\omega)\) from \(\omega = 0.1\) rads\(^{-1}\) to \(\omega = 42.2\) rads\(^{-1}\) for the different SPPH (full SAOS spectra in Figure S13). The mechanical percolation thresholds \(\phi_{\text{NP}}\) of the NPs of the SPPH at different \(n_l\) (Figure 3C) are obtained by fitting the data to a classic percolation model, \(G'(\omega) = K(\phi - \phi_{\text{c}})\), where \(K\) is a prefactor and \(\gamma\) is a scaling exponent related to the dimensionality of the percolating system.\(^3,5,9\) As hypothesized, the NP percolation threshold \(\phi_{\text{NP}}\) is correlated to linker concentration \(n_l\) in excellent agreement with the micrographs showing that low values of \(r\) drive local anisotropy in NP self-assembly and low-density SPPH morphologies (Figure 2B,C). The relationship between the scaling exponent \(\gamma\) and \(n_l\) tells a similar story (Figure 3D). All obtained exponents are below \(\gamma \approx 2\), indicating that NP
Percolation in SPPH is analogous to percolation of resistor networks or gels with isotropic forces,\textsuperscript{39,41} in good agreement with prior studies on polymer-bridged NPs and PEG networks.\textsuperscript{36,40} Importantly, $\gamma$ also varies systematically with $n_l$ and decreases from $\gamma \approx 2.0$ to $\gamma \approx 1.2$ as the linker concentration is lowered from $n_l = 15.0\%$ to $n_l = 1.8\%$. This trend is indicative of a progressive reduction in the dimensionality of the percolating system from 3-D to values close to 2-D (per numerical simulation estimates, $\gamma \approx 1.5-2.0$ for 3-D and $\gamma \approx 1.1-1.3$ for 2-D),\textsuperscript{36,39-41} in excellent agreement with the expected onset of anisotropy at lower linker concentrations and $r$ (Figure 2A–C). These results confirm that controlling the NP interaction valency provides programmable anisotropy, morphology, and percolation threshold of the SPPH, thus corroborating theoretical predictions\textsuperscript{2,6,8} and advancing our ability to design structurally versatile biomimetic patchy particle systems.

Lastly, we demonstrate a strategy to control the self-assembly and percolation behaviors of the SPPH independently from the aforementioned method of varying linker concentration $n_l$. Specifically, we hypothesized that introducing transient cross-linking species to compete with the NPs for the 4nPEG linkers may reduce the number of linkers that form between NPs, such that the asymptotic increase in $r$ with increasing $N_{4nPEG}$ at low $\phi_{NP}$ (Figure 3A) may be avoided. Our hypothesis is motivated by patchy particle assembly in biological settings, which often takes place in crowded viscoelastic environments such as the cellular cytosol which have many competing weak interactions.\textsuperscript{8,42,43} Thus, to mimic such a crowded competitive macromolecular environment for the SPPH, we introduce $\text{Fe}^{3+}$ ions to the 4nPEG linkers ($n_l = 10$ wt $\%$) and NP mixture. In this configuration, the $\text{Fe}^{3+}$ ions will compete with the $\text{Fe}^{3+}$ ions on the $\text{Fe}_3\text{O}_4$ NP surface for the nitrocatechol groups in 4nPEG (thus reducing $r$), and form transient cross-links with the 4nPEG linkers via coordination bonds to form a viscoelastic fluid (Figure 4A). We refer to this system as a supramolecular patchy particle hydrogel with competing interactions (SPPH-CI).

Figure 4. Independent control of the valency of particle–particle interactions in the SPPH ($n_l = 10$ wt $\%$) by introducing competing interactions. (A) Schematic of the crowded SPPH with competing ion interactions (SPPH-CI). The $\text{Fe}^{3+}$ ions introduced in the gel compete with the $\text{Fe}_3\text{O}_4$ NPs for the 4nPEG linkers to form difunctional coordination complexes (red box). (B) Representative SEM micrographs of the SPPH-CI system at $\phi_{NP} = 0.75\%$ at different magnifications. Top panel scale bar = 100 $\mu$m; bottom panel scale bar = 10 $\mu$m. Anisotropic striations are observed throughout the sample, which are also observed in the $\phi_{NP} = 0.45\%$ gel and in different samples of the $\phi_{NP} = 0.75\%$ gel (Figure S16). (C) Evolution of the storage modulus $G'(\omega)$ (solid symbols) and loss modulus $G''(\omega)$ (open symbols) of the SPPH-CI with increasing $\phi_{NP}$ at 4nPEG concentrations of $n_l = 10$ wt $\%$ (see Figure S18C for additional $\phi_{NP}$ values). The data are obtained from SAOS measurements at 5 °C (circles) and 25 °C (diamonds) and shifted to a reference temperature of 25 °C. Solid and dashed lines indicate fits of the dynamic moduli to the FZM. The insets in the $\phi_{NP} = 0.00\%$ and $\phi_{NP} = 0.75\%$ panels show representative images of the SPPH-CI system at the respective NP concentrations, demonstrating a stark transition from a mobile viscoelastic liquid to a rubbery gel. Scale bar = 1 cm. (D) Elastic contributions of the $\text{Fe}^{3+}$ ion network and the $\text{Fe}_3\text{O}_4$ NP network to the overall viscoelasticity of the SPPH-CI as a function of $\phi_{NP}$ based on FZM predictions (Figure S18D,E).
(Figures 4B and S16). This shows that the SPPH-CI system is able to retain the sheet-like layers of NPs—previously observed in the regular SPPH via TEM (Figure 2C)—at mesoscopic length scales due to the presence of the transiently cross-linked ion network. We also observe regular porosity in the microstructure of the SPPH-CI samples, which appears to gain prominence with increasing $\phi_{NP}$ (Figure S14) and can be attributed to NP-rich domains. Lastly, TEM micrographs of the diluted SPPH-CI show clear evidence of assortative self-assembly44 leading to a clear separation of NP-rich and ion-rich domains (Figure S15), justifying the proposed microstructure of the SPPH-CI (Figure 4A).

The onset of these highly anisotropic structures is also correlated with a much earlier onset of mechanical percolation of the NP network in the SPPH-CI compared to the regular SPPH, as demonstrated by rheological measurements of the SPPH-CI as a function of $\phi_{NP}$ for $n_i = 10.0\%$ (Figure 4C). Evidence of a contribution to the elastic modulus from the USAXS (Figure S17). The obvious separation of time scales for the ion and NP relaxations in Figure 4C allows us to capture the individual contributions of the ion and NP networks to the viscoelastic moduli of the 4nPEG + Fe$^{3+}$ transient network (Figures 4B and S16). This shows that the SPPH-CI system is able to retain the sheet-like layers of NPs—previously observed in the regular SPPH via TEM (Figure 2C)—at mesoscopic length scales due to the presence of the transiently cross-linked ion network. We also observe regular porosity in the microstructure of the SPPH-CI samples, which appears to gain prominence with increasing $\phi_{NP}$ (Figure S14) and can be attributed to NP-rich domains. Lastly, TEM micrographs of the diluted SPPH-CI show clear evidence of assortative self-assembly44 leading to a clear separation of NP-rich and ion-rich domains (Figure S15), justifying the proposed microstructure of the SPPH-CI (Figure 4A).

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CONCLUSION

We have presented a supramolecular polymer-bridged patchy particle hydrogel system that can serve as an experimental platform for designing particle gels which not only are phase-stable but also offer tunability in the particle–particle interaction valency, thus providing access to a wide range of morphologies and percolation behaviors that have been predicted by theory. We show that the valency of the particle–particle interaction can be tuned by adjusting the number of polymer linkers per particle and also independently by adding competing interactions to the system. The precise quantitative relation between $r$ and the coordination number $M$ remains unclear; however, we anticipate that additional simulations would provide critical insight into the M of the SPPH and facilitate greater rational control over the design of the anisotropic supramolecular hydrogels. Beyond our SPPH system, we expect the findings in this work to be salient for a wide range of gel systems with controllable cross-link functionalities, such as DNA gels,24 metal–organic cage gels,33 protein gels,8,11 bridged particle gels,36,45–51 and multifunctional polymer gels.52 We anticipate these findings will accelerate the design of bioinspired programmable soft materials for structural and biomedical applications and inspire deeper studies of the physics of patchy particle self-assembly in diverse environments.

METHODS

Polymer Synthesis. Unless specifically stated otherwise, all chemicals are purchased from Sigma-Aldrich. The 4-arm PEG N-hydroxysuccinimide (10 kDa, JenKem Technology) was functionalized with nitrocatechol using a variation of previously reported methods to create 4-arm nitrocatechol PEG (4nPEG).53 Nitroaniline end-groups were first synthesized by dissolving 1.00 g of dopamine hydrochloride and 1.45 g of sodium nitrite in 30 mL of water. The reaction vessel was then placed into an ice bath, and 5.0 mL of 20% w/w sulfuric acid was slowly added under vigorous stirring. The yellow precipitate was collected through vacuum filtration, washed with cold water once and cold methanol three times, and vacuum-dried. The 4-arm PEG N-hydroxysuccinimide was then functionalized with the collected nitroaniline salt by dissolving 1.0 g of 4-arm PEG N-hydroxysuccinimide and 0.24 g of the nitroaniline salt (2x equivalent of N-hydroxysuccinimide) in 12 mL of DMF. The reaction vessel was then moved to an oil bath at 55 °C, and 0.2 mL of TEA (>2x equivalent of N-hydroxysuccinimide) was added to initiate the coupling. The reaction was left to proceed under N$_2$ overnight. Afterward, the product was extracted with chloroform and dried using sodium sulfate. The product was then precipitated in diethyl ether, three times and vacuum-dried. The functional efficiency of the 4nPEG was typically around ~88% (see Calculations section in the Supplementary Information).

The 1-arm PEG N-hydroxysuccinimide (2 kDa, JenKem Technology) and 4-arm PEG N-hydroxysuccinimide (10 kDa, JenKem Technology) were functionalized with catechol to create 1-arm catechol PEG (1cPEG) and 4-arm catechol PEG (4cPEG) following the same method but without the conversion step from dopamine to nitroaniline.

Nanoparticle Synthesis. Oleylamine-stabilized Fe$_3$O$_4$ NPs with an average diameter of 7 nm were synthesized by reductive decomposition of an iron salt in a heat-up method.44,45 In a typical synthesis, 1.05 g of iron(III) acetylaconetate (99%, Strem Chemicals) was dissolved in a mixture of 15 mL of benzyl ether (99%, Acros Organics) and 15 mL of oleylamine (98%) in a 100 mL, three-necked, round-bottomed flask. Oleylamine serves as both the reducing agent
and stabilizing ligand. The mixture was vigorously stirred and degassed under vacuum at room temperature for 1 h, followed by heating (ramp rate of \( \sim 7 ^\circ C/min \)) at 120 \( ^\circ C \) for 1 h. During the temperature ramp, the flask was backfilled with nitrogen at 90 \( ^\circ C \). The mixture was then quickly heated under inert atmosphere to 210 \( ^\circ C \) (heating rate of \( \sim 15 ^\circ C/min \)) and held for 1 h. Upon cooling to 50 \( ^\circ C \), 50 mL of ethanol was added to the dark brown reaction product, followed by subsequent centrifugation and redispersion in THF. The oleylamine-functionalized \( \text{Fe}_3\text{O}_4 \) NPs dispersed in THF were then functionalized with 1cPEG by adding 1cPEG to the mixture at a 1cPEG/\( \text{Fe}_3\text{O}_4 \) mass ratio of 3:1. The mixture was heated to 55 \( ^\circ C \), and ligand exchange was allowed to proceed for 24 h. The resulting solution was then precipitated in hexane three times before being dried under vacuum.

**SPPH Gel Synthesis.** SPPH gels are made by mixing aqueous solutions of 4nPEG, 1cPEG-NP, and HCl to make a final volume of 200 \( \mu L \) and pH of \( \sim 2.2 \), with varying amounts of 4nPEG and 1cPEG-NP depending on the \( n_t \) and \( \phi_{NP} \) (note that \( \phi_{NP} \) is defined with respect to the volume of the \( \text{Fe}_3\text{O}_4 \) core, not the entire 1cPEG-NP complex).

An acidic environment for the SPPH is chosen due to the fact that a pH of \( \sim 2 \) facilitates stable mono-coordinations between nitrocatechol iron and the \( \text{Fe}_3\text{O}_4 \) surface.\(^{58} \) At higher pH of \( \sim 5.2 \), where affinities are stronger and bis-coordinations dominate,\(^{58} \) we found that the NPs showed some signs of etching, manifested by an emergence of an \( \text{Fe}^{3+} \) ion relaxation mode even in the absence of \( \text{Fe}^{3+} \) ions added (Figure S20). Therefore, an acidic environment is used to prevent any trace amounts of \( \text{Fe}^{3+} \) ions in the 1cPEG-NP solution from participating in the cross-linking of the gel. SPPH with competing ions (SPPH-CI) are made by mixing aqueous solutions of 4nPEG, 1cPEG-NP, \( \text{FeCl}_3 \) (added with a stoichiometric ratio of 2 nitrocatechol to 1 \( \text{Fe}^{3+} \) )), and NaAc buffer to form a final volume of 200 \( \mu L \) and a pH of \( \sim 5.2 \) to facilitate bis-coordinations (Figure 4A). Although the etching effect found in the regular SPPH at this pH will likely contribute to an increased availability of \( \text{Fe}^{3+} \) beyond the stoichiometric amount added, this does not affect the gelation of the system, and the etching effect on the size of the NPs was negligible, as indicated by USAXS scattering intensities of the SPPH-CI compared to the regular SPPH (Figure S17 and Table S1). For a typical sample, the mixture is sealed and placed in a 55 \( ^\circ C \) oven for 24 h to create a gel.

**Nuclear Magnetic Resonance.** \(^1\)H spectra of 4nPEG were obtained using a Varian 300 MHz spectrometer on a 4nPEG sample dissolved in CDCl\(_3\) (10 mg/mL).

**Dynamic Light Scattering (DLS)**. DLS measurements were made using an Anton-Paar Litesizer 100 particle size analyzer. The number-weighted size distribution of 4nPEGs (Figure S1B) was obtained on much more diluted samples (0.01 wt %) due to the black opaque color of the specimen. Measurements were repeated three times to ensure reproducibility of data.

**Ultrasmall-Angle X-ray Scattering.** USAAXS and SAXS intensities were obtained using the USAAXS instrument at 9-1D beamline at the Advanced Photon Source (APS). Samples were mounted in cells with a sample thickness of \( \sim 1 \) mm and with 3M Scotch tape on either side. Due to variations in the thickness of the actual samples, only relative intensities are reported in the paper. X-ray energy was 21 keV (wavelength = 0.5905 Å), and the beam size was 0.6 × 0.6 mm for USAAXS and 0.1 × 0.6 mm for SAXS. Data collection was 90 s for USAAXS and 15 s for SAXS. Data were subtracted from a blank cell with Scotch tape, reduced, and then merged together using software (Indra, Nika, and Irena) provided by the beamline.\(^{59,60} \) Slit smearing inherent to the USAAXS instrument was removed using desmearing routines in Irena to generate pinhole equivalent data for analysis.

**Time-Dependent Small-Angle X-ray Scattering.** Time-dependent SAXS measurements were carried out using a pinhole camera at the small-angle XPCS beamline 8-ID-D at the APS. Samples were mounted in cells with a sample thickness of 1.5 mm and were sealed with thin polycarbonate windows on either side. Due to variations in the thickness of the actual samples, only relative small-angle scattering intensities are reported in the paper. Monochromatic X-rays at an energy of 7.35 keV were used, and the beam size was 15 \( \mu m \times 15 \mu m \), with a nominal flux of 10\(^9\) photons/s, which is then attenuated by a factor of 14. A large area medipix based detector array (LAMBDA) with a pixel size of 55 \( \mu m \) and array size of 516 \( \times \) 1556 was used. The measurements were checked for radiation damage by controlling the dose using filters, and an optimum dose was determined for the measurements.

**Linear Rheology.** All viscoelastic measurements of the hydrogels were conducted using Anton-Paar’s MCR-302 stress-controlled rheometer, with a 10 mm parallel plate geometry and a gap size of 1 mm. All gels were loaded, trimmed, and sealed with mineral oil prior to characterization to prevent dehydration. Time-dependent small-amplitude oscillatory shear (SAOS) experiments for gelation measurements in Figure 1F were conducted at an angular frequency \( \omega = 5 \text{ rads}^{-1} \) at an amplitude of \( \gamma = 1\% \). All other SAOS experiments were conducted at an amplitude of \( \gamma = 1\% \). Step strain experiments for relaxation measurements in Figure 1G were conducted at an amplitude of \( \gamma = 1\% \). Large amplitude oscillatory shear measurements were conducted at an angular frequency \( \omega = 5 \text{ rads}^{-1} \) to ensure that \( \gamma = 1\% \) is within the linear regime (Figure S4C).

## ASSOCIATED CONTENT

### Supporting Information

The Supporting Information is available free of charge at https://pubs.acs.org/doi/10.1021/acsnano.0c06389.

Fitting parameters, calculation and analysis schemes, NMR/DLS characterization of polymers and nanoparticles, and supplementary rheological data, scattering curves, and microscopy images of the SPPH and SPPH-CI systems (PDF)

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Notes
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