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Regulating Volume Transitions of Highly Responsive Hydrogel Scaffolds by Adjusting the Network Properties of Microgel Building Block Colloids

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We present a simple method to control the volume change of thermally responsive hydrogel scaffolds, providing a remarkably fast swelling and deswelling response to temperature changes. These scaffolds have 3-dimensional colloidal-network structures which are made from microgel particles while they are above their deswelling transition temperatures. By tuning the cross-link density of the microgel particles, we achieve controllable changes of the volume of the scaffolds in response to temperature. Their fast response rate is determined by the length scale of the unit microgel particles and is not influenced by the properties of the network. The release profile of a model drug (Rhapontin) loaded within the scaffolds can also be regulated by the cross-linking density of the microgel particles. These results offer a new way of fabricating hydrogel scaffolds with tunable matrix geometry and function by adjusting the properties of the unit microgel colloids, without loss of their fast response to temperature change.

Introduction

Stimuli-responsive hydrogels can swell or deswell in response to external triggers such as pH,[1] temperature,[2] electric field,[3] light,[4−7] or biomolecules.[8] Such hydrogels have many potential practical applications including drug delivery,[9−12] nanopatterning,[13] chemical and biosensing,[14,15] and photonic crystals.[16−18] In all these applications, it is highly desirable that the hydrogels respond quickly to the applied stimulus. For example, a fast response allows the matrix properties to be switched to control a drug release pattern while minimizing a hysteresis. Fast response has usually been achieved by incorporating additional porosity into the hydrogel,[19−21] by grafting dangling chains onto the hydrogel,[22−24] or by hybridizing nanoparticles within the polymer networks.[25,26] However, it remains difficult to achieve fast response with these techniques if the length of the hydrogels remains macroscopic, since the relaxation of length of a cross-linked hydrogel network is controlled by diffusion of liquid and is hence proportional to the square of its dimension.[27,28]

In a previous study, we have overcome this limitation by fabricating a novel hydrogel scaffold system that used submicrometer microgel building block particles assembled into a 3-dimensional network. The microgel particles aggregated to form the scaffold while heated above their transition temperature (T_{tr}) through bridging or depletion interactions, and the scaffold was then covalently bonded by chemical means.[29] This resultant scaffold exhibits a remarkably fast response to temperature changes, while also being able to easily immobilize biomolecules and functional colloids within the network of microgel particles.

To extend the application of such scaffold systems, it is necessary to find a suitable means of regulating the functions of the scaffold, such as degree of swelling and drug release behavior. The degree of swelling for most conventional hydrogels is controlled by changing their cross-linking density, but this also changes their response kinetics.[30,31] Thus, it remains a challenge

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(1) Tanaka, T. *Phys. Rev. Lett*. **1978**, *40*, 820–823.
(2) Hirokawa, Y.; Tanaka, T. *J. Chem. Phys*. **1984**, *81*, 6379–6380.
(3) Kwon, I. C.; Bae, Y. H.; Kim, S. W. *Nature* **1991**, *354*, 291–293.
(4) Suzuki, A.; Tanaka, T. *Nature* **1990**, *346*, 345–347.
(5) Juodkazis, S.; Mukai, N.; Wakaki, R.; Yamaguchi, A.; Matsuo, S.; Misawa, H. *Nature* **2000**, *408*, 178–181.
(6) Ikeda, T.; Nakano, M.; Yu, Y.; Tsutsumi, O.; Kanazawa, A. *Adv. Mater*. **2003**, *15*, 201–205.
(7) Lendlein, A.; Jiang, H.; Junger, O. *Nature* **2005**, *434*, 879–882.
(8) Miyata, T.; Asami, N.; Uragami, T. *Nature* **1999**, *399*, 766–769.
(9) Pelton, R. *Adv. Colloid Interface Sci*. **2000**, *85*, 1–33.
(10) Lindman, S.; Lynch, I.; Thulin, E.; Nilsson, H.; Dawson, K. A.; Linse, S. *Nano Lett*. **2007**, *7*, 914–920.
(11) Zhang, Y.; Guan, Y.; Zhou, S. *Biomacromolecules* **2007**, *7*, 3196–3201.
(12) Reese, C. E.; Mikhonin, A. V.; Kamenjicki, M.; Tikhonov, A.; Asher, S. A. *J. Am. Chem. Soc*. **2004**, *126*, 1493–1496.
(13) Koholek, M.; Lee, W. K.; LaMattina, B.; Caster, K. C.; Zauscher, S. *Nano Lett*. **2004**, *4*, 373–376.
(14) Kim, J.; Nayak, S.; Lyon, L. A. *J. Am. Chem. Soc*. **2005**, *127*, 9588–9592.
(15) Debord, J. D.; Lyon, L. A. *J. Phys. Chem. B* **2000**, *104*, 6327–6331.
(16) Hu, Z.; Huang, G. *Angew. Chem., Int. Ed*. **2003**, *42*, 4799–4802.
(17) Nyak, S.; Lee, H.; Chmielewski, J.; Lyon, L. A. *J. Am. Chem. Soc*. **2004**, *126*, 10258–10259.
(18) Gorelikov, I.; Field, L.; Kumacheva, E. *J. Am. Chem. Soc*. **2004**, *126*, 15938–15939.
(19) Dong, L. C.; Hoffman, A. S. *J. Controlled Release* **1990**, *13*, 12–31.
(20) Zhang, X. Z.; Yang, Y. Y.; Chung, T. S.; Ma, K. X. *Langmuir* **2001**, *17*, 6094–6099.
(21) Kuang, M.; Wang, D.; Gao, M.; Hartmann, J.; Mohwald, H. *Chem. Mater*. **2005**, *17*, 656–660.
(22) Yoshida, R.; Uchida, K.; Kaneko, Y.; Sakai, K.; Kikuchi, A.; Sakurai, Y.; Okano, T. *Nature* **1995**, *374*, 240–242.
(23) Kaneko, Y.; Nakamura, S.; Sakai, K.; Aoyagi, T.; Kikuchi, A.; Sakurai, Y.; Okano, T. *Macromolecules* **1998**, *31*, 6099–6105.
(24) Xu, X.-D.; Zhang, X.-Z.; Yang, J.; Cheng, S.-X.; Zhuo, R.-X.; Huang, Y.-Q. *Langmuir* **2007**, *23*, 4231–4236.
(25) van Durme, K.; van Mele, B.; Loos, W.; du Prez, F. E. *Polymer* **2007**, *46*, 9851–9862.
(26) Petit, L.; Bouteiller, L.; Brulet, A.; Lafuma, F.; Hourdet, D. *Langmuir* **2007**, *23*, 147–158.
(27) Tanaka, T.; Fillmore, D. J. *J. Chem. Phys*. **1979**, *70*, 1214–1218.
(28) Matsuo, E. S.; Tanaka, T. *J. Chem. Phys*. **1988**, *89*, 1695–1703.
(29) Cho, E. C.; Kim, J.-W.; Fernandez-Nieves, A.; Weitz, D. A. *Nano Lett*. **2008**, *8*, 169–172.
(30) Cima, L. G.; Lopina, S. T. *Macromolecules* **1995**, *28*, 6787–6794.
(31) Barbucci, R.; Magnani, A.; Consumi, M. *Macromolecules* **2000**, *33*, 7475–7480.

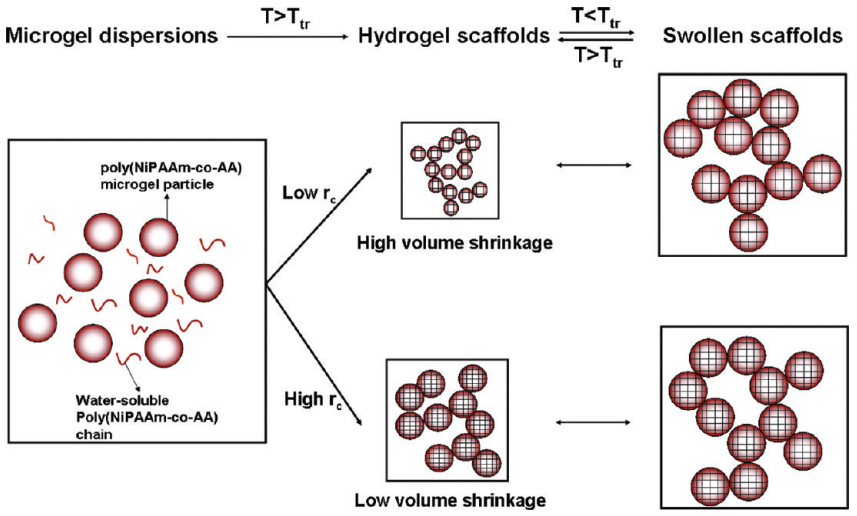

Figure 1. Schematic showing the preparation of hydrogel scaffolds by using microgel particles of poly(*N*-isopropylacrylamide-*co*-allyamine), poly(NiPAAm-*co*-AA). The as-prepared microgel particle dispersion contains some water-soluble poly(NiPAAm-*co*-AA) chains, with different chemical compositions from the microgel particles or with short chain lengths enough for them to be solublilized in water. Because of the depletion force and hydrophobic interaction between microgel particles, heating the microgel particles dispersion to above the transition temperatures of the microgel particles (T_{tr}) makes the microgel particles form clusters, allowing us to produce 3-dimensional hydrogel scaffolds. The scaffold volume at $T > T_{tr}$ can be regulated by using microgel particles of different cross-link densities, by varying r_c. At $T < T_{tr}$, all scaffolds have similar volumes. The microgel particles have similar diameters (~0.7 μm) and surface charges (~+20 mV). Once the scaffolds are formed, they retain their shapes after repeated heating and cooling.

to create hydrogel scaffolds with controllable properties which retain a fast response to changes in temperature.

Here we report a simple method to control the swelling and deswelling volumes of hydrogel scaffolds without deterioration of their fast response to changes in temperature. The approach is shown schematically in Figure 1. We use submicrometer-sized microgel particles of different cross-link densities to fabricate hydrogel scaffolds; this regulates the properties of the scaffolds. Since the length scale of the unit microgel particles is responsible for the fast response of the scaffolds, we can make scaffolds that retain a similar fast response to temperature changes despite having different properties. We demonstrate how the unit microgel particles affect the physical properties of the hydrogel scaffolds, such as volume changes and swelling and deswelling kinetics. We also demonstrate that the drug release behavior of the scaffolds is influenced by the network properties of the microgel particles.

Experimental Section

Materials. *N*-Isopropylacrylamide (NiPAAM), allyamine (AA), potassium persulfate (KPS), and *N,N,N,N*-dimethylene bis(acrylamide) (BIS) were obtained from Aldrich. Methacryloxythiocarbonyl rhodamine B (Polyfluor 570) and Rhapontin (Rh) were purchased from Polysciences and Kaden Biochemicals GmbH, respectively.

Synthesis of Microgel Particles. We dissolved 4 g of NiPAAM, 139 μL of AA, 0.12 g of KPS, and the cross-linker (BIS) in 160 mL of deionized water and heated this solution to 70 °C. After 1 h polymerization, the dispersion was rapidly cooled to 4 °C. While cooling, we sonicated (Branson 1500, 70 W and 42 kHz output frequency) the dispersion to prevent aggregation of the microgel particles. We stored the microgel dispersions at 4 °C before use. We label these dispersions "as-prepared". The cross-link density of the microgel particles was regulated by varying the molar ratio of BIS to NiPAAM (r_c). We prepared three representative samples with r_c of 0.037, 0.075, and 0.11. We also prepared dye-labeled microgel particles the same way, except 2 mg of Polyfluor 570 was added to the monomer mixture solution before polymerization. After dialyzing the as-prepared dispersions in deionized water for 3 days and then freeze-drying them, we determined the concentration of microgel particles to be 1.4−1.8 wt % in the dispersions.

Preparation of Hydrogel Scaffolds. We used the as-prepared microgel particle dispersions for the preparation of hydrogel scaffolds. In addition to the microgel particles of poly(*N*-isopropylacrylamide-*co*-allyamine), poly(NiPAAM-*co*-AA), the as-prepared dispersions could also contain some water-soluble poly(NiPAAM-*co*-AA), unreacted monomers, and initiators. 10 mL of as-prepared microgel particle dispersions (1.4 wt %) was stored in an oven at 65 °C for 4 h to produce scaffolds. We removed unreacted monomer and initiator by repeatedly washing, swelling, and reheating the scaffolds. The resultant scaffolds were stored at 4 °C before use. The present work differs from our previous studies[29] in that we did not use chemical glues such as glutaldehyde to fix the scaffolds. Drug-containing hydrogel scaffolds were prepared by using the same process with the addition of 1 wt % of a hydrophobic drug, Rh, to the dispersion of microgel particles just before heating the sample to 65 °C.

Characterization of Microgel Particles and Hydrogel Scaffolds. The structures of microgel particles and hydrogel scaffolds were observed with a confocal microscope (Axiovert 200 M with an LSM 510 Laser Module, Carl Zeiss). The sizes of microgel particles were measured with photocorrelation spectroscopy (PCS, Malvern Instruments 4500HS) in the temperature range 20−40 °C. Prior to the measurement, the as-prepared microgel dispersions were dialyzed for 3 days to remove unreacted monomer, initiator, and water-soluble polymer. The surface charges of the microgel particles were measured at room temperature with the same instrument. The temperature-dependent volume changes of the hydrogel scaffolds were determined by measuring the length of the scaffolds after they were equilibrated in 200 mL of deionized water at a given temperature for 2 h. The deswelling kinetics of the hydrogel scaffolds was investigated by measuring the dimensions of the scaffolds after applying a thermal shock: we transferred the scaffolds stored in deionized water at 20 °C to vessels containing deionized water at 45 °C. We recorded the dimensional changes of the scaffolds with time by analyzing movies taken during the experiment. We observed the thermal behavior of the microgel particles and the scaffolds by using differential scanning calorimetry (DSC, DSC Q1000, TA

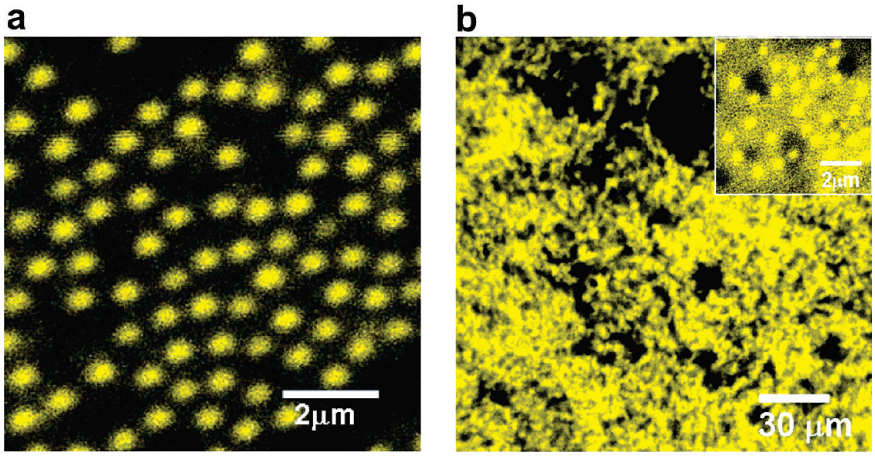

Figure 2. (a) A confocal microscope image of microgel particles with $r_c = 0.037$. (b) Microstructure of a hydrogel scaffold prepared from the microgel particles with $r_c = 0.037$. Inset is a close-up of the microstructure, showing that the scaffold consists of microgel particles.

Instruments). The scaffolds or microgel particles contained in a sample pan were scanned from 0 to 70 °C with a heating rate of 1 °C/min. The microstructures of the scaffolds equilibrated at either room temperature or 40 °C were observed by scanning electron microscopy (SEM, Hitachi S4200) after the quenching the scaffolds to −70 °C and freeze-drying them.

Drug Release Behavior of Hydrogel Scaffolds. Rh-loaded hydrogel scaffolds ($r_c = 0.037$ and 0.075) were preincubated in deionized water at 20 °C for 30 min before use. The scaffolds were rapidly transferred to glass containers filled with 200 mL of deionized water at room temperature (22 °C) and body temperature (37.5 °C), respectively. We observed the release profile of Rh from the scaffolds by monitoring the concentration of Rh in the aqueous solution for 72 h with a high-performance liquid chromatograph (Hewlett-Packard 1100). We sampled 1 mL of solution at a certain intervals, and we filled the samples with the same amount of deionized water to keep the volume of the solution constant. The mobile phase was a mixture of water/acetonitrile (7/3, v/v) with 0.1 wt % H_3PO_4, the flow rate was 1 mL/min, and the column was a Mightysil C18 (4.6 × 250 mm, 5 μm). The concentration of Rh was determined by the area of the peak detected at 324 nm. We accounted for any loss of drugs due to purification and pre-equilibration of the scaffolds to precisely compare the release profiles between the scaffolds. Three independent runs were conducted for the statistical treatment of the data.

Results and Discussion

Volumetric and Thermal Transitions of the Hydrogel Scaffolds. We have previously reported that hydrogel scaffolds can be made by simply heating thermo-responsive microgel particles.[29] This approach ensures that the small length scales of the microgel particles govern the time behavior of the resultant scaffold.[27,28] In the current study, we show that this technique also enables us to control the temperature-responsive volume changes of the hydrogel scaffolds by adjusting the cross-link density of the microgel particles. To demonstrate this, we fabricate hydrogel scaffolds using microgel particles of different cross-link densities, regulated by varying the molar ratio of BIS to NiPAAM, r_c. We synthesize poly(NiPAAm-*co*-AA) microgel particles which are 0.76 ± 0.02, 0.70 ± 0.02, and 0.72 ± 0.01 μm in diameter for the r_c of 0.037, 0.075, and 0.11, respectively. The particles are stably dispersed in water without any aggregates below T_{tr} (35 °C), as shown in Figure 2a. Prior to the synthesis of scaffolds, we store the as-prepared microgel dispersion at below 10 °C to prevent any possible aggregation of the particles.

As shown in Figure 1, the as-prepared microgel particle dispersion also contains some water-soluble poly(NiPAAm-*co*-AA) chains which are generated during the synthesis of microgel particles. These linear polymers could have different chemical compositions from the poly(NiPAAm-*co*-AA) microgel particles or could be short in their lengths enough to be solublilized in water. We take advantage of these polymers to construct the hydrogel scaffolds. Because of the combination of depletion force and hydrophobic interaction between microgel particles, heating the as-prepared microgel particles dispersion to above T_{tr} makes the microgel particles form clusters, allowing us to produce 3-dimensional hydrogel scaffolds. Once the scaffolds are formed, the scaffolds retain their shapes without using chemical glue even if they have experienced repeated heating and cooling cycles. We speculate that the microgel colloids in the scaffolds can be physically cross-linked via intermolecular entanglement between particles, and this entanglement is mediated by water-soluble polymers. We have found that the microgel dispersions do not form hydrogel scaffolds, even when stored for long times above T_{tr}, after the water-soluble polymers are removed from the microgel dispersions by dialysis for 3 days. This result further demonstrates that the water-soluble polymers play a critical role in the formation of hydrogel scaffolds. Confocal microscopy analysis (Figure 2b) shows that the scaffold is highly porous, consisting of microgel particles randomly connected to one another. On the basis of these results, the formation of the hydrogel scaffolds is irreversible; once formed, they are not disassembled into individual particles in water, even when temperature goes back below T_{tr}. The scaffolds can swell to the initial reaction volume below T_{tr}, but most microgel particles are not isolated and form a permanent structure.

By regulating the cross-linking densities through control of r_c, we make scaffolds of different volumes at $T > T_{tr}$. We observe that a higher degree of cross-linking of the microgel particles leads to a lower volume shrinkage of the scaffold at 65 °C. The volume of the scaffold decreased as r_c became lower (Figure 3). However, when the temperature decreases below T_{tr}, the volumes of the scaffolds is essentially the same, independent of the value of r_c. During repeated heating and cooling cycles, the volume of the scaffold remains the same at a given temperature, indicating that the swelling and deswelling of the scaffolds is a fully reversible process. We plot the dimensions of three scaffolds as a function of temperature in Figure 4a. The scaffold with the lowest r_c shows the biggest change in size around T_{tr}, as compared with the other scaffolds with higher r_c. Interestingly, this trend is the same as that of the corresponding microgel particles, as shown in Figure 4b. This result implies that the volumetric transitions of the scaffolds were controlled by the behavior of their unit microgel particles.

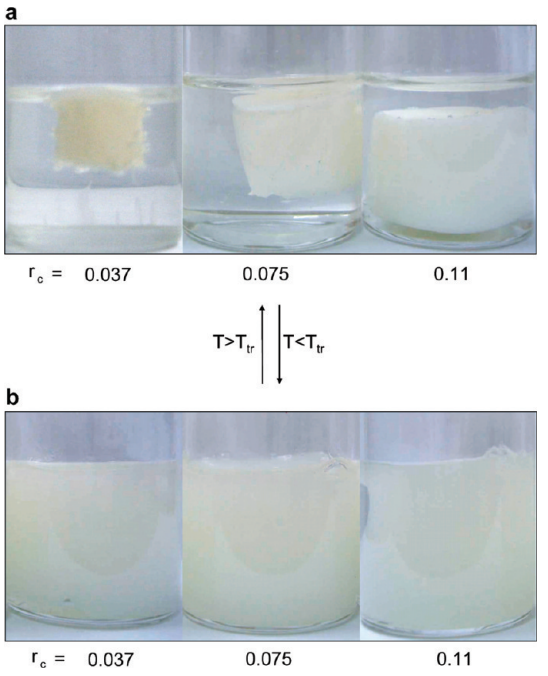

Figure 3. Photographs of hydrogel scaffolds (a) at 65 °C and (b) at room temperature. The scaffolds are made from microgel particles of different cross-link densities, r_c.

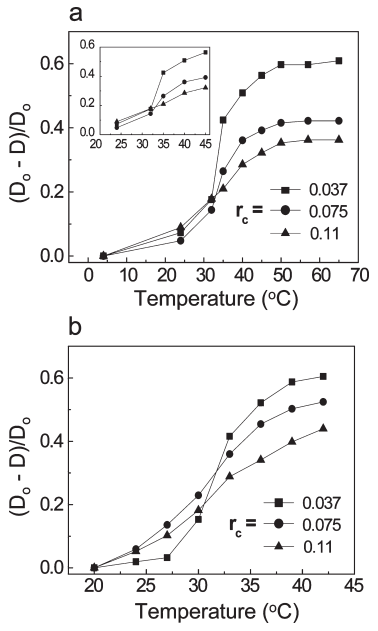

Figure 4. Relative changes in dimension, $(D_0 - D)/D_0$, of (a) hydrogel scaffolds and (b) microgel particles as a function of temperature. D is the length of the scaffolds or the diameter of the microgel particles at a given temperature, and D_0 is the initial length of the scaffolds at 4 °C or the diameter of the microgel particles at 20 °C. Inset in (a) shows the data in the same temperature range as shown in (b).

The use of microgel building block particles of different r_c results in big differences in the capabilities of the hydrogel scaffolds to swell and deswell in response to temperature changes. However, such a difference in the volumetric behavior could deteriorate their fast response rates.[23,30,31] To investigate this possibility, we determine the relaxation times of the three hydrogel

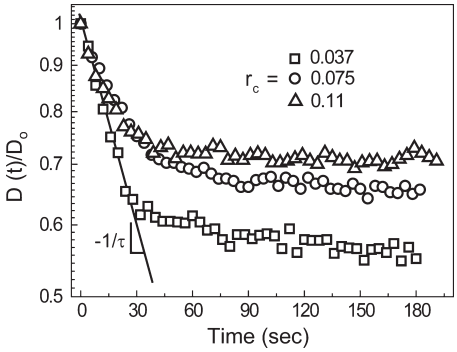

Figure 5. Temporal evolution of the swelling ratio, $D(t)/D_0$, for hydrogel scaffolds fabricated using microgel particles with different r_c. D_0 is the initial sizes of the scaffolds. From the exponential fits shown by the solid lines, we obtain the characteristic deswelling times (τ) for the hydrogel scaffolds. To experimentally determine the deswelling kinetics, we quenched the scaffolds by transferring them from a water vessel at 20 °C to one at 45 °C.

scaffolds by measuring the temporal evolution of their dimensions upon transferring the scaffolds stored at 20 °C to water vessels at 45 °C. The results are shown in Figure 5. We obtain the relaxation times, τ, of the three scaffolds, where τ characterizes the decay time of the size: $D(t) = (D_i - D_f)e^{-t/\tau} + D_f$.[32,33] Here, $D(t)$ is the scaffold diameter at time t, D_i is the initial size, and D_f is the final size. The values of τ are 64, 95, and 94 s for $r_c = 0.037$, 0.075, and 0.11, respectively. Interestingly, the values of τ for these three scaffolds are not much different, and they all show responses that are 2 orders of magnitude faster than those of bulk hydrogel. The τ of conventional bulk hydrogels is usually $\sim 1.5 \times 10^4$ s.[33,34] The diffusion coefficients of three hydrogel scaffolds obtained from τ_s and using the equation in ref 27 are also similar ($(1-1.5) \times 10^{-4}$ cm^2/s), which are much higher than those of bulk hydrogels (3.2×10^{-7} cm^2/s).[27,33,34]

Remarkably, the cross-link density of the microgel particles has little effect on the response kinetics of the resultant scaffolds. For the three hydrogel scaffolds, we use microgel particles with nearly identical particle sizes ($\sim$0.7 μm), surface charges ($\sim$+20 mV), and polymer concentrations in the dispersion (1.4 wt %), and there is no significant difference in the network structures between the scaffolds. Under these conditions, the response kinetics of the three scaffolds should be the same because unit microgel particles, with similar length scales, give the scaffolds similar relaxation times.

To further prove that the responses of the hydgrogel scaffolds are mainly governed by their unit microgel particles, we compare the thermal properties of the individual particles with those of the corresponding hydrogel scaffolds, measured using DSC (Figure 6). The three scaffolds had similar T_{tr}, 33.5−34 °C, which were quite close to those of the microgel particles at 35 °C, as shown in Figure 6a,b. However, they showed different heats for their phase transitions; the scaffold with a lower r_c needs more heat, which is likely due to a larger volume change. More importantly, Figure 6c shows that there is only a small difference between the heats for the transition of the microgel particles and the corresponding scaffolds ($\sim$5 J/g_{gel}). This result demonstrates that that the thermal transition of the scaffolds mostly originate from the transition of the microgel particles themselves, thereby

(32) Okajima, T.; Harada, I.; Nishio, K.; Hirotsu, S. *J. Chem. Phys.* **2002**, *116*, 9068–9077.
(33) Li, Y.; Wang, G.; Hu, Z. *Macromolecules* **1995**, *28*, 4194–4197.
(34) Bansil, R.; Liao, G.; Falus, P. *Physica A* **1996**, *231*, 346–358.

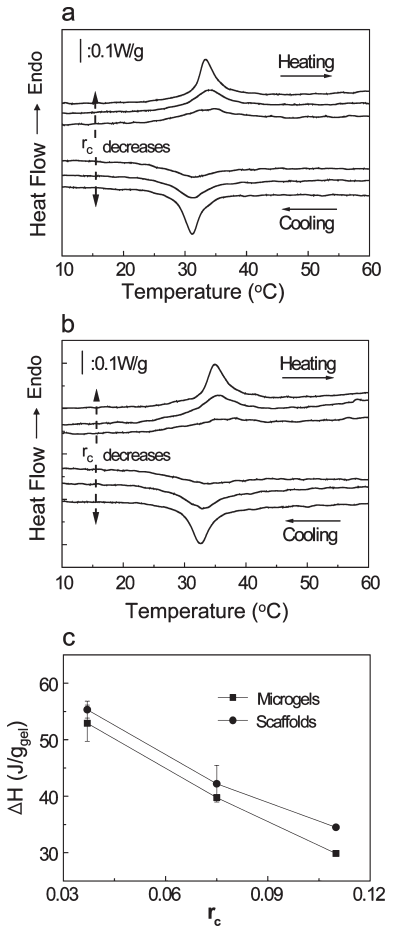

Figure 6. (a) DSC thermograms for hydrogel scaffolds fabricated using microgel particles of different r_c: 0.037, 0.075, and 0.11. (b) DSC thermograms for the microgel particles. Cooling experiments were carried out after finishing a heating scan with the same scan rate. (c) Transition heats for the microgel particles and the hydrogel scaffolds as a function of r_c.

allowing us to conclude that the volume change of the hydrogel scaffolds are regulated by the unit microgel particles.

Drug Release Behavior of the Hydrogel Scaffolds. To investigate how the network properties of microgel particles influence the drug release behavior of hydrogel scaffolds, we load a hydrophobic drug, Rh, into the hydrogel scaffolds. This drug is a rhubarb root component with an inhibitory effect on tyrosinase activity.[35] To investigate the effect of r_c, we fabricate hydrogel scaffolds with Rh using microgel particles with r_c = 0.037 and 0.075. We monitor the release behavior of Rh from the scaffolds at body temperature (37.5 °C) and at room temperature (22 °C). The T_{tr} of the scaffolds lies between these temperatures. At body temperature (Figure 7a), Rh is rapidly released from the scaffolds; the released amount reaches a plateau in ~1 h. By contrast, at room temperature (Figure 7b), Rh is also released rapidly in the early stages but does not show any plateau. A comparison of the release behavior at the two temperatures (see insets) reveals that the release rate of Rh at body temperature during the first 60 min was higher, ~0.0005 min^{-1}, than that at room temperature, ~0.0002 min^{-1}. However, the total amount of Rh released by 72 h was much higher at room temperature. The faster release in

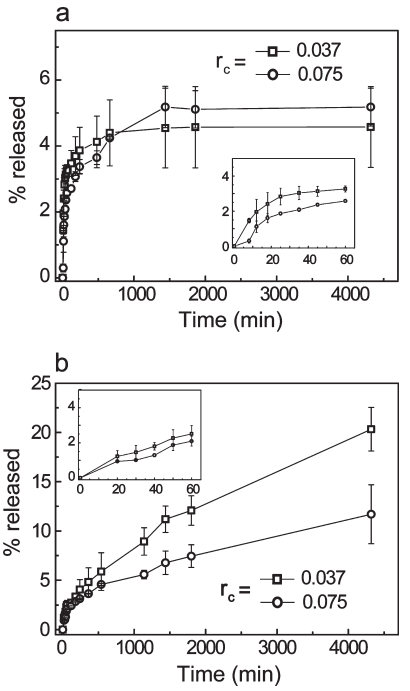

Figure 7. Release profiles of Rhapontin loaded in hydrogel scaffolds prepared from microgel particles of different r_c, 0.037 and 0.075: (a) at body temperature (37.5 °C); (b) at room temperature (22 °C). Insets show release profiles of Rhapontin in the scaffolds during the first 60 min.

the early stage at body temperature might be due to fast shrinkage of the scaffold, thereby depleting the drug molecules on the surface.[36] After the fast release, the drug within the scaffold stop being released, mainly due to the small volume and dense structure formed resulting from the shrinkage of the scaffold, which limited the diffusion of drug molecules. In addition, hydrophobic interaction between the scaffolds and hydrophobic drug could also limit the drug-release from the scaffolds.[37] By contrast, at room temperature, the drug could readily diffuse out even after the initial fast release due to the opening of the diffusion pathways in the network.

Two scaffolds showed no significant differences in the release rate and total amount of release at body temperature, while the amount of drug released from the scaffold with r_c = 0.037 at room temperature was 2 times higher than the scaffold with r_c = 0.075. To elucidate the release behavior of the two scaffolds, we checked their microstructures at different temperatures by SEM (Figure 8). From the images, we see that the microstructures of the scaffolds at the two temperatures are clearly different; the scaffolds are more densely packed at 40 °C than at room temperature. However, for any given temperature, the microstructures of the two scaffolds are not much different. This suggests that the difference in the drug-release behavior of the two scaffolds at room temperature can be attributed to the properties of the microgel particles, rather than the microstructure or porosity of the scaffolds. In particular, at body temperature, the release rate and the amount of drug released are roughly the same because the two scaffolds show a similar temperature response, regulated by the unit microgel particles. At room temperature, where the drug release is controlled by diffusion, microgels with higher r_c result in more sustained release of drugs

(35) Kim, Y. M.; Yun, J.; Lee, C.-K.; Lee, H.; Min, K. R.; Kim, Y. *J. Biol. Chem.* **2002**, *277*, 16340–16344.

(36) Gutowska, A.; Bark, J. S.; Kwon, I. C.; Bae, Y. H.; Cha, Y.; Kim, S. W. *J. Controlled Release* **1997**, *48*, 141–148.

(37) Choi, C.; Chae, S. Y.; Nah, J.-W. *Polymer* **2006**, *47*, 4571–4580.

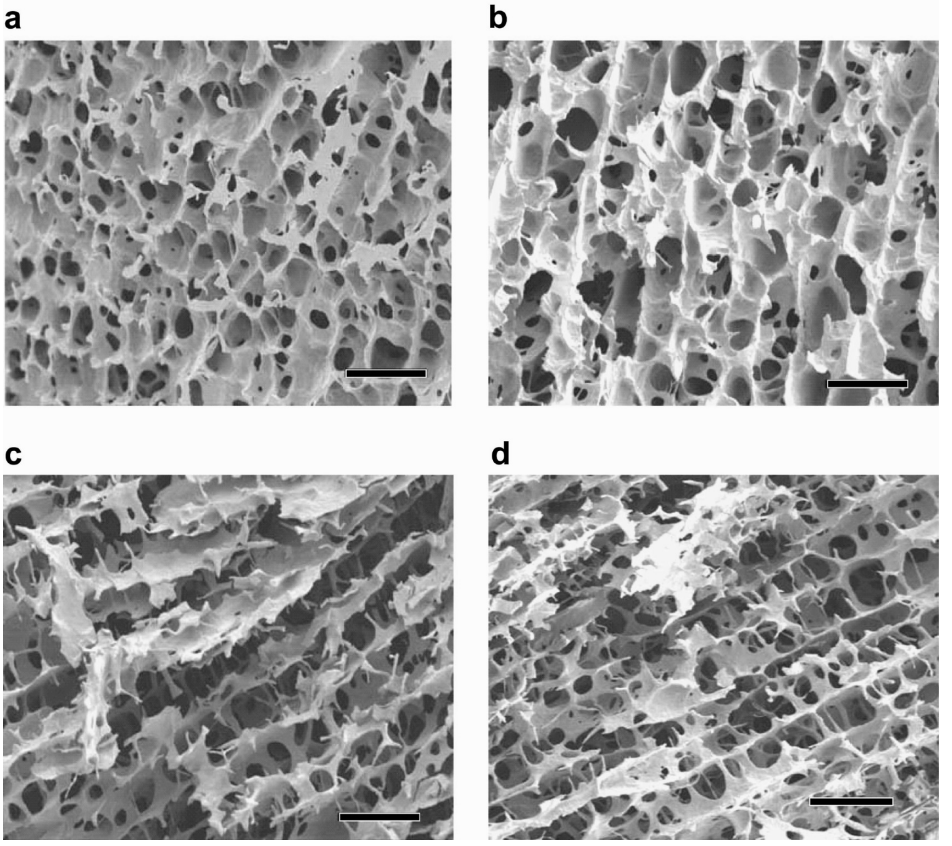

Figure 8. SEM images of hydrogel scaffolds for $r_c = 0.037$ (a, c) and 0.075 (b, d). These scaffolds were prepared by freeze-drying the quenched (to −70 °C) scaffolds, which were previously equilibrated at either 40 °C (a, b) or room temperature (c, d). Scale bar is 50 μm.

from hydrogel networks.[38−40] These results suggest that by controlling r_c of the unit microgel particles, we can control the release of drugs from hydrogel scaffolds, while still maintaining a fast response at a targeted temperature.

Conclusion

We present a method to control the volume transition of hydrogel scaffolds fabricated from submicrometer-sized microgel particles. By changing the cross-linking density of the unit microgel particles, we can control the volumetric properties of the hydrogel scaffolds while still maintaining their fast responses to temperature changes. In addition, we also find that the properties of the microgel particles determine the release profile of a drug loaded in the scaffolds. Thus, we can regulate the properties of the hybridized materials under controlled conditions, while also quickly switching their functions by external stimulus. This approach should be useful and practical for constructing a variety of smart delivery systems.

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(38) Wu, L.; Brazel, C. S. *Int. J. Pharm.* **2008**, *349*, 144–151.
(39) Young, S.; Wong, M.; Tabata, Y.; Mikos, A. G. *J. Controlled Release* **2005**, *109*, 256–274.
(40) Eichenbaum, G.; Kiser, P. F.; Dobrynin, A. V.; Simon, S. A.; Needham, D. *Macromolecules* **1999**, *32*, 4867–4878.