



**Construction of a nano-phase-separated structure on a hydrogel surface**

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## COMMUNICATION

## Construction of a nano-phase-separated structure on a hydrogel surface

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**A hydrogel surface with a nano-phase-separated structure was successfully fabricated by grafting a fluorine-containing polymer using activators regenerated by electron transfer atom transfer radical polymerisation (ARGET ATRP). The modified hydrogel surface exhibits water repellency and high elasticity with maintaining transparency.**

Polymer hydrogels are composed almost entirely of water and a three-dimensional (3D) cross-linked polymer network. They have unique properties, such as holding large amounts of water in their networks, acting as open systems that can freely exchange both matter and energy with their surroundings, and their elastic moduli are similar to those of biological substances. Therefore, these hydrogels are used in a wide variety of applications, such as drug delivery carriers, artificial cartilages, cell culture scaffolds, and flexible sensors.<sup>1–4</sup> To date, hydrogels have been considered bulk materials in many studies, and methods for designing chemical and network structures of whole hydrogels have been developed.<sup>5–9</sup> However, it has been reported that not only the bulk structures of hydrogels but also their surfaces have a large influence on their properties.<sup>10–12</sup> Given the essential roles played by hydrogel surfaces in determining hydrogel properties, there has been an increase in the number of studies on functionalising the hydrogel surfaces, e.g. attaching an elastomer to a hydrogel surface<sup>13,14</sup>, grafting a polymer to a hydrogel surface<sup>15–17</sup>, and grafting a polymer from a hydrogel surface<sup>18–21</sup>. Although extensive research efforts have been devoted to developing functionalisation methods for hydrogel surfaces, the nanoscale morphologies of the modified hydrogel surfaces have been poorly discussed. Herein we report a novel method for

fabricating a nanoscale phase-separated structure on a hydrogel surface. In this method, a fluorine-containing polymer was grafted from the initiators of atom transfer radical polymerisation (ATRP) immobilised on the surface of the hydrogel. An intrinsic limitation of the morphological study of hydrogel surfaces is the difficulty in their characterisation. In many studies, modifications of the hydrogel surfaces were proved by several analyses, in which the resolution in the horizontal direction is up to the micrometre scale, such as FTIR measurements or fluorescence imaging using dye molecules.<sup>20,21</sup> Here, we successfully visualised the nanoscale structure on the hydrogel surface by adhesion force mapping of AFM measurements using hydrophobised tips for the first time. Then, we carried out some demonstrations to investigate the influence of the nanoscale structure on the hydrogel properties. In particular, the hydrogel surface exhibits water repellency despite domain-like modifications. This study has the potential to lead to further functionalisation of hydrogels by utilising their surfaces.

To fabricate the nano-phase-separated structure on the hydrogel surface, a hydrogel with immobilised ATRP initiators on the surface region was prepared according to a previous report.<sup>18</sup> First, a non-grafted hydrogel (NG gel) was synthesised by redox radical polymerisation of *N,N*-dimethylacrylamide (DMAAm), with 3-(*N*-aminopropyl)methacrylate (NAPMAm) as an initiator immobilisation site and *N,N'*-methylenebis(acrylamide) (MBAAm) as a crosslinker. The ATRP initiators were immobilised onto the surface of the NG gel by immersing it in an aqueous solution of the ATRP initiators with an activated ester moiety for a short time and then dialysed against water to remove the unreacted ATRP initiators. The initiator-immobilised hydrogel (IG gel) was replaced with dimethylformamide (DMF), and 1H,1H,5H-octafluoropentylacrylate (OFPA) was polymerised from the immobilised ATRP initiators by activators regenerated by electron transfer (ARGET) ATRP using CuBr<sub>2</sub>, tris[2-(dimethylamino)ethyl]amine (Me<sub>6</sub>TREN), and ascorbic acid as the copper source, ligand, and reducing agent, respectively. After polymerisation, the hydrogel was dialysed against DMF to remove the unreacted monomer and gradually replaced with water. The obtained fluorine-containing polymer-grafted hydrogel is abbreviated as FG gel (Fig. 1).

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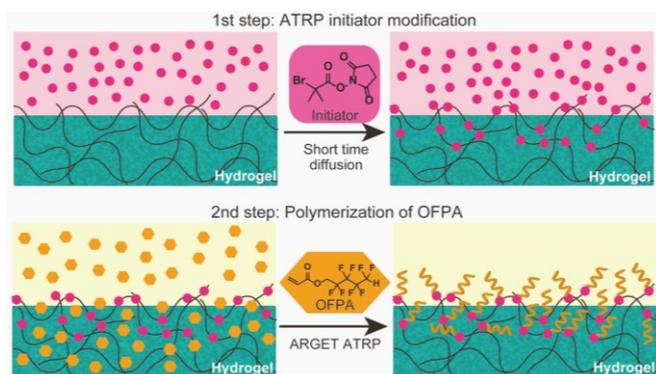


Fig. 1 Schematic illustration of surface modification of hydrogels

To prove the introduction of poly(1H,1H,5H-octafluoropentyl acrylate (POFPA) on the surface region of the FG gel, IR spectra of FG and IG gels were obtained using an attenuated total reflection Fourier transform infrared (ATR/FT-IR) spectrometer (Fig. 2a). Although the IR spectral shapes of both the IG gel and the cross-section of the FG gel almost correspond to the water, the IR spectrum of the surface region of the FG gel shows several significant peaks assigned to the C-F vibration ( $1130\text{ cm}^{-1}$  and  $1169\text{ cm}^{-1}$ ) of OFPA. In other words, IR measurements revealed that C-F vibration signal of OFPA was confirmed only on the surface region of the FG gel. Furthermore, confocal Raman microscopy revealed the depth profiles of POFPA in FG gel. From the Raman spectra of POFPA, NG gel, and FG gel, the peak from C=O vibration was appropriate for estimating the amount of POFPA at a certain depth (Fig. S2). Fig. 2b shows the intensity profiles of C=O vibration ( $1760\text{ cm}^{-1}$ ) in the NG gel and FG gel in the range of 1–1000  $\mu\text{m}$  from their surfaces. The intensity of the NG gel at  $1760\text{ cm}^{-1}$  was almost the same at all depths. In contrast, the intensity of the FG gel at  $1760\text{ cm}^{-1}$  decreased as the distance from the surface increased and reached zero at 200  $\mu\text{m}$ . These results indicate that POFPA was successfully introduced into the surface region of the FG gel.

Next, atomic force microscopy (AFM) measurements were carried out to analyse the surface morphology of the NG and FG gels in water. Adhesion force mapping was conducted on the NG and FG gel surfaces using hydrophobic or hydrophilic tips. The adhesion force images of hydrogels using hydrophobic tips revealed that 10–100 nm of domain-like high adhesion areas were formed only on the FG gel surface (Fig. 3, S4). The histograms of the adhesion force on the FG gel surface observed by both hydrophobic and hydrophilic tips showed bimodal distributions (Fig. 3f,h), although the histograms of the NG gel surface show unimodal distributions (Fig. 3b,d). These results clearly indicate that hydrophobic and hydrophilic phases coexist on the FG gel surface. These results suggest that grafted POFPA forms a hydrophobic nano-sized phase-separated structure on the FG gel surface, and the hydrophobic tip visualised the difference in hydrophobicity between these two phases. Maps of height and elasticity also drastically changed after the grafting of POFPA (Fig. S3). The surface elasticity of the NG gel is increased from  $108.2 \pm 19.1$  to  $492.4 \pm 108.3\text{ kPa}$  ( $n=16$ ) by the surface modification. The roughness of NG gel and FG gel were  $8.84 \pm 4.28$  and  $2.47 \pm 0.28\text{ nm}$ , respectively. These results suggest that the surface region of the FG gel collapsed because of the hydrophobic interactions between the POFPA chains. We hypothesised that the introduced hydrophobic

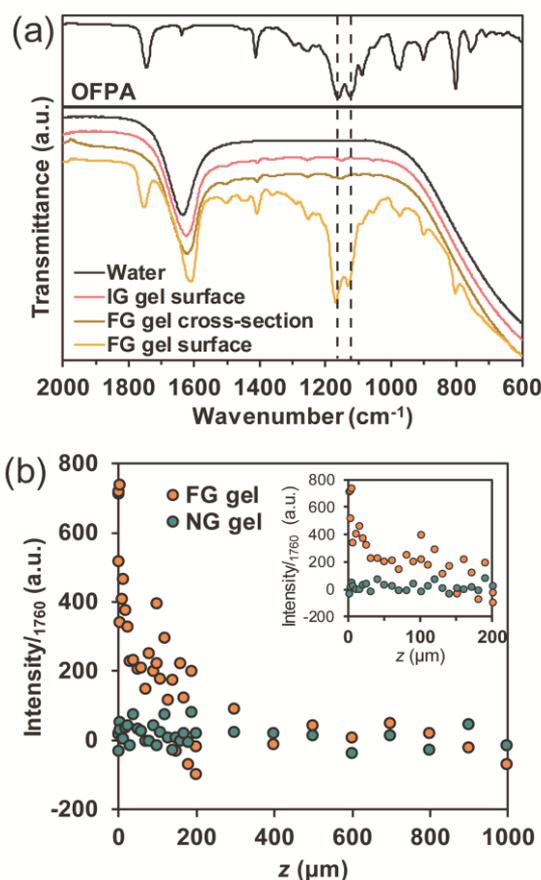


Fig. 2 Characterisation of the surface regions of IG gel and FG gel. (a) IR spectra of OFPA, IG gel and FG gel. (b) Depth profiles of C=O vibration peak from the surface of IG and FG gel. Inset: Closer look of 0–200  $\mu\text{m}$  region from the surface.

domain deswells the hydrogel network, and the density of the polymer increased at the surface region. It should be noted that the FG gel was almost transparent in the visible light region (optical transparency > 90%, 400–700 nm) even when the hydrophobic polymer chains were introduced (Fig. 4). In general, hydrophobic part in hydrogels is likely to form aggregates and the aggregation makes hydrogels opaque due to the scattering of visible light.<sup>22,23</sup> In this case, we hypothesised that a fixed POFPA chain could not form large aggregates and nanoscale domains were formed. Consequently, the high optical transparency of the FG gel was maintained because the phase-separated structure was sufficiently small compared with the wavelength of visible light.

To evaluate the influence of the nano-phase-separated surface on the hydrogel properties, contact angle measurements were carried out. The water contact angle after 120 s from dropping water on the FG gel surface ( $91.8^\circ \pm 0.9^\circ$ ) is much higher than that on the NG gel ( $39.0^\circ \pm 1.9^\circ$ ) and IG gel ( $57.7^\circ \pm 1.4^\circ$ ), as shown in Fig. 5. And the contact angle of *n*-hexadecane on FG gel surface ( $44.9^\circ \pm 4.3^\circ$ ) is also higher than that on the NG gel ( $18.8^\circ \pm 4.4^\circ$ ), as shown in Fig. S6(a). These results indicated that the hydrophobic domain structure introduced on the surface affected the wettability of the hydrogel.

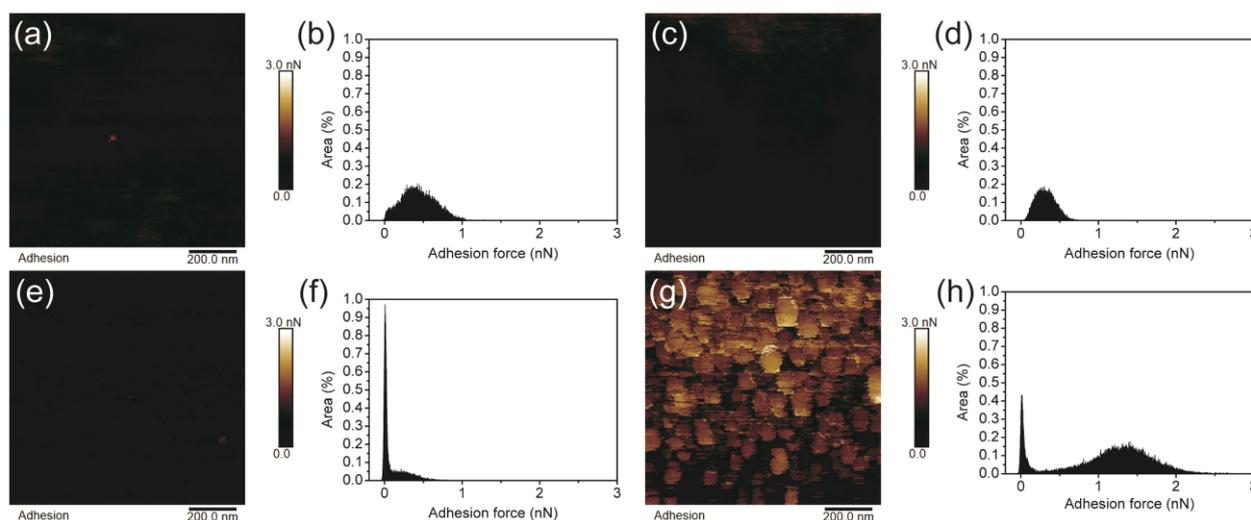


Fig. 3 Maps and histograms of adhesion force on NG gel with a hydrophilic tip (a,b) and a hydrophobic tip (c,d); FG gel with a hydrophilic tip (e,f) and a hydrophobic tip (g,h).

We successfully demonstrated the introduction of a nano-phase-separated structure on the hydrogel surface. The nano-phase-separated surface was prepared on the hydrogel by surface-initiated ARGET ATRP of OFPA. The introduction and depth profile of the POFPA-grafted chain was confirmed by ATR/FT-IR and confocal Raman microscopy. The phase-separated structure on the hydrogel surface was confirmed by adhesion force mapping using AFM for the first time. The nano-phase-separated structure did not interfere with the transparency of the hydrogel. In contrast, the grafted POFPA on the hydrogel surface had a large influence on the wettability and elasticity of the FG gel surface. This study provides a new strategy to modify surface morphologies of hydrogels by the post-synthetic approach. The strategy can be applied to versatile hydrogel systems instead of the PDMAAm hydrogel which we demonstrated. Furthermore, the size and the density of the phase-separated

structure might be controlled by changing the grafting conditions. We envision that the size-dependent properties of hydrogel surfaces, such as a permeability, can be tuned in our future work. Our methodology could shed the light on the design of hydrogel surfaces with nanostructures

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## Conflicts of interest

There are no conflicts to declare.

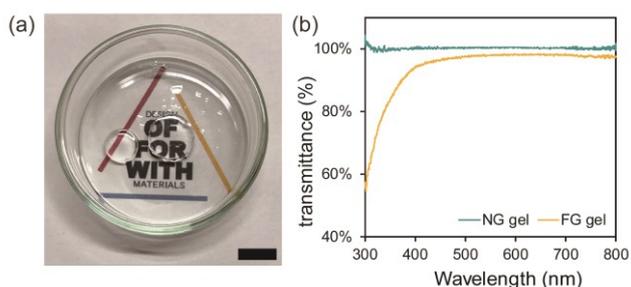


Fig. 4 (a) Image of FG gel (left) and NG gel (right). Scale bar = 1 cm. (b) Transmittance spectra of NG and FG gel.

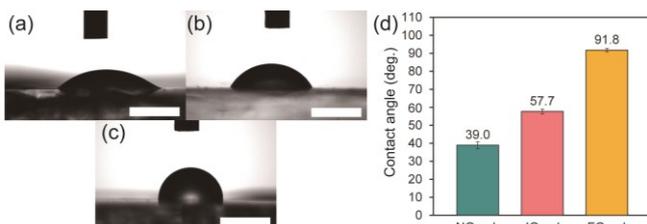


Fig. 5 Images of water drop deposited on (a) NG gel, (b) IG gel, and (c) FG gel. Scale bars = 1 mm. (d) The contact angles of a water drop.

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