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Smart Hydrogels Co-switched by Hydrogen Bonds and π–π Stacking for Continuously Regulated Controlled-Release System

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A series of hydrogels with continuously regulatable release behavior can be achieved by incorporating hydrogen bonding and π–π stacking co-switches in polymers. A poly(nitrophenyl methacrylate-*co*-methacrylic acid) hydrogel (NPMAAHG) for control over drug release is fabricated by copolymerizing 4-nitrophenyl methacrylate and methacrylic acid using ethylene glycol dimethacrylate as a crosslinker. The carboxylic acid groups and nitrylphenyl groups form hydrogen bonds and π–π stacking interactions, respectively, which act as switches to control the release of guest molecules from the polymers. As revealed by the simulated gastrointestinal tract drug release experiments, the as-synthesized NPMAAHG hydrogels can be regulated to release only 4.7% of drugs after 3 h in a simulated stomach and nearly 92.6% within 43 h in the whole digestive tract. The relation between the release kinetics and structures and the mechanism of the smart release control are analyzed in terms of diffusion exponent, swelling interface number, drug diffusion coefficient, and velocity of the swelling interface in detail. The results reveal that the release of guest molecules from the hydrogels can be continuously regulated for systemic administration by controlling the ratio of the hydrophilic hydrogen bonds and the hydrophobic π–π stacking switches.

1. Introduction

Hydrogels have attracted great attention because of their smart response to specific changes in environmental conditions.[1–10] Controlled-release drug delivery is one of the promising applications of smart gels.[4,11–14] The smart behavior of hydrogels is generally based on non-covalent dynamic bonding interactions, e.g., hydrogen bonding,[1–3] hydrophobic,[8] π–π stacking,[9,10] and electrostatic interactions,[11,14] which respond to stimuli such as pH,[2,7] temperature,[3,7] ultraviolet[6] or visible lights,[8] chemical substances[1] or electric field,[11,14] respectively. In this work, we incorporate both hydrogen-bonding and π–π stacking interactions in polymer hydrogels to achieve a dual switched on–off control over drug release for oral administration.

Oral administration is the most convenient route of chemotherapy.[15,16] Drugs for oral administration need protection against the hostile acidic environment and high concentrations of proteolytic and metabolytic enzymes in the stomach, and their release needs to be controlled to fit the pharmacokinetics. Poly(methacrylic acid) (PMAA) as a well known controlled release hydrogel works on the basis of hydrogen bonding between carboxylic acid groups (COOH) in the crosslinking network. The hydrophilic nature of PMAA limits the ability to effectively load hydrophobic and large macromolecular drugs.[17–19] The introduction of hydrophobic units can increase the load of hydrophobic drugs and reduce the toxic burst release effects.[20–24] Currently used hydrophobic units are usually long-chain alkyl groups, hydrophobic esters, or cyclodextrins which form hydrophobic domains by van der Waals forces with a lower interaction energy. Incorporation of hydrophobic segments overcomes the issue of loading hydrophobic drugs, but it is still a problem to have a long sustained controlled drug release that corresponds to the transit time in the gastrointestinal tract (3–4 h in stomach and 36–48 h for whole digestive tract).[25,26]

The π–π stacking interaction is a strong physical interaction between aromatic groups within a distance of 3.645–3.928 Å.[9,10,23] The π–π stacking interactions formed by *N*-(fluorenyl-9-methoxycarbonyl)-amino (Fmoc-amino) acids can cooperate with short peptide derivatives to form a fibrous hydrogel for application in biological sensing.[27] Other substances such as phenylalanine (Phe)[28] and modified *N*-fluorenylmethoxycarbonyl-amino acids[10,29] are used for cell culture and tissue engineering applications. The key role that aromatic interactions play the

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biological field lie in their free energy of formation of an ordered and directional structure as well as good biocompatibility. Herein, we pioneer a strategy of incorporating π–π stacking interactions in a PMAA hydrogel system to achieve a co-switched controlled release of drugs that is consistent with the transit time in the gastrointestinal tract. The stronger physical interactions between the aromatic groups performed as π–π stacking switches retain the release of drugs and consequently change the drug release kinetics. The hydrophobic nature of the π–π stacking interactions can provide a hydrophobic domain for storage of hydrophobic drugs. The π–π stacking interactions are introduced using 4-nitrophenyl methacrylate (NPMA), which is synthesized by reacting acyl chloride with 4-nitrophenol. Then NPMA is copolymerized with methacrylic acid (MAA) using ethylene glycol dimethacrylate (EGDMA) as crosslinker to prepare a poly-(nitrophenyl methacrylate-*co*-mathacrylate acid) hydrogel (NPMAAHG). Simulated gastrointestinal tract drug release experiments are carried out to investigate the drug delivery property of NPMAAHG by using ibuprofen (IBU) as a model drug. Scanning electron microscopy (SEM), UV-vis spectroscopy, and X-ray diffraction (XRD) are used to investigate the π–π stacking interactions and the crystallographic nature of the as-synthesized hydrogels. In addition, the pH sensitivity and drug release kinetics properties of the as-prepared hydrogels are also examined.

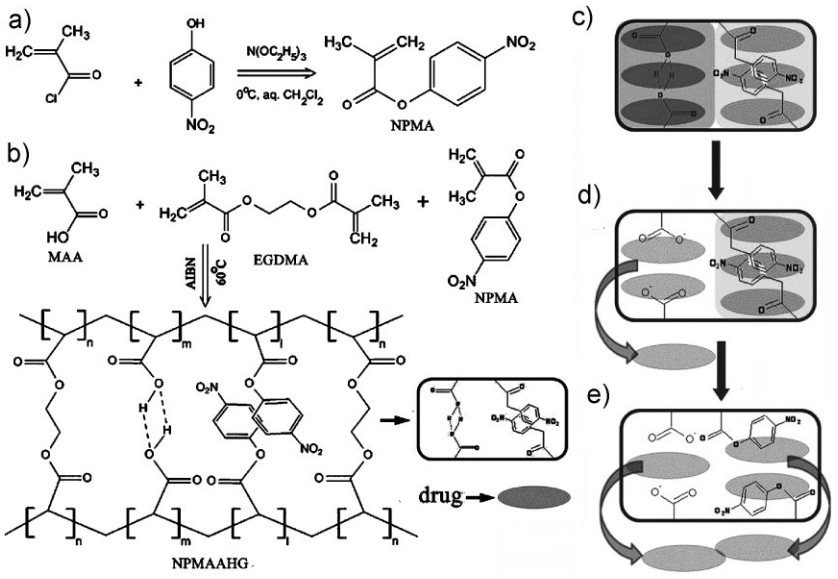

Figure 1. Schematic illustration of the preparation, structure, and co-switched drug release mechanism of the NPMAAHG hydrogels. a) Synthesis of the functional comonomer NPMA. b) Copolymerization of NPMA with MAA to obtain the NPMAAHG hydrogel by using EGDMA as a crosslinker. The carboxylic acid groups and NPMA segments form hydrogen bonding and π–π stacking interactions, respectively, which act as switches to control the release of guest molecules. c) Drugs are closed in NPMAAHG at low pH (1.4); hydrogen bonding and π–π stacking interactions perform as off-status switches to retard the release of drugs. d) Drugs partially release from the NPMAAHG networks owing to the opening of the hydrogen bond switch in basic solution; the π–π stacking switch still remains. e) Drugs further release owing to the opening of the π–π stacking switch caused by the swelling of the hydrogels.

2. Results and Discussion

2.1. Design and Preparation of Functional Hydrogels

To achieve a long sustained control over drug release suitable for systematic administration, hydrophobic aromatic groups with strong interaction forces are incorporated into the EGDMA crosslinked network of a PMAA hydrogel. Figure 1 displays the synthetic route used to prepare the targetted hydrogels. First, a functionalized comonomer NPMA is synthesized by modifying 4-nitrophenol with methacryloyl chloride (Fig. 1a). The functionalized monomer is then copolymerized with MAA using EGDMA as a crosslinker to prepare a NPMAAHG hydrogel (Fig. 1b). The carboxylic acid groups and NPMA segments in NPMAAHG form hydrogen bond and π–π stacking interactions, respectively, which act as switches to control the release of guest molecules (Fig. 1c). π–π stacking interactions between NPMA segments cover an effective area of 7.74×3.96 Å (Fig. S1 in the Supporting Information). Crosslinked by EDGMA, the distance between two neighboring alkyl chains is confined to ≈10.79 Å. Considering the size of ibuprofen molecules as 10.29×5.24 Å,[30] the structure of NPMAAHG ensures the storage and maintenance of drug molecules effectively. In the high pH (7.4) solution, the ionization of the carboxylic acid groups leads to the invalidity of the hydrogen bond switch, but the π–π stacking switch still remains. Thus, drugs are partially released at the initial stage (Fig. 1d). The ongoing swell of the hydrogel disables the π–π stacking interactions, and drugs are further released (Fig. 1e). In the low pH (1.4) solution, the hydrogen bonds and π–π stacking interactions act as physical crosslinks to resist the release of guest molecules from their network (Fig. 1c). The polymerization is carried out in ordinary test tubes under protection of N_2 for about 24 h at 60 °C. After the product is soaked in a water/acetone (50:50 v/v) solvent mixture for one week, a colorless and transparent hydrogel of NPMAAHG is obtained (Fig. 2). By changing the molar ratios of NPMA and MAA from 1:2, to 1:4, to 1:8, NPMAAHGs are synthesized and noted as NPMAAHG-2, NPMAAHG-4, and NPMAAHG-8, respectively.

2.2. Drug Release Behavior of NPMAAHG Hydrogels

Figure 3 displays the simulated gastrointestinal release of IBU from NPMAAHG-4, which is synthesized with a NPMA-to-MAA molar ratio of 1:4. The drug storage of the hydrogels is 33.45% (386.87 mg of ibuprofen per gram of NPMAAHG-4) loaded in 40 mg mL^{-1} IBU solution. Drug-loaded NPMAAHG-4 is first placed in simulated gastric fluid (SGF) (pH 1.4) for 3 h, the transit time of substances in the stomach. It is then transferred into simulated intestinal fluid (SIF) (pH 7.4) until all drugs are released

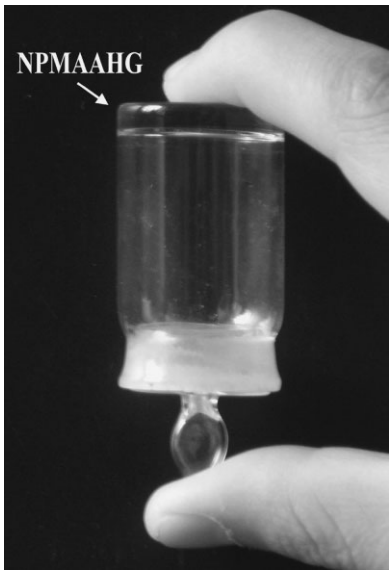

Figure 2. Image of the NPMAAHG hydrogels. The hydrogel in the bottom of the bottle (note bottle is inverted) shows a transparent colorless nature.

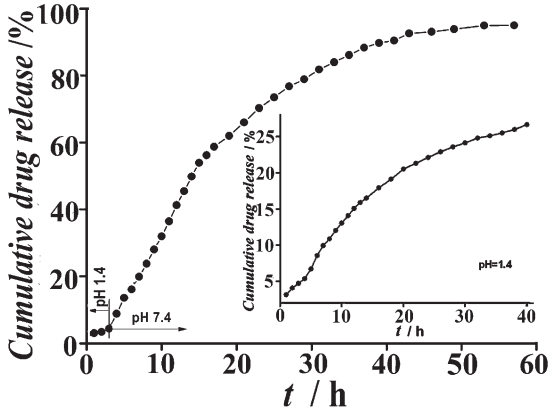

Figure 3. Release of IBU from the NPMAAHG-4 hydrogel system by simulating the gastrointestinal tract. After 3 h incubation in SGF at pH 1.4, the drug-loaded NPMAAHG-4 hydrogel is transferred to SIF at pH 7.4 until all drugs are released. Inset: drug release in SGF at pH 1.4.

from the system at 37 °C. Only 4.7% of the IBU molecules are released during the first 3 h in SGF, and then the drugs are released increasingly after transferring the hydrogel from the SGF to the SIF solution. It takes 43 h in the SIF solution to achieve 92.6% drug release and 48 h to achieve 98% drug release, which accords well with the transit time through the gastrointestinal tract. The slow drug release in the SGF (pH 1.4) is presented as an insert in Figure 3. Therefore, by introducing functional NPMA segments, we have a successful control over the drug delivery during the transit through the gastrointestinal tract.

The drug release rate from NPMAAHG can be regulated by varying the ratio of the hydrogen bonds to π–π stacking interactions. Figure 4 presents the in vitro drug release curves of IBU-loaded NPMAAHG-2, NPMAAHG-4, and NPMAAHG-8

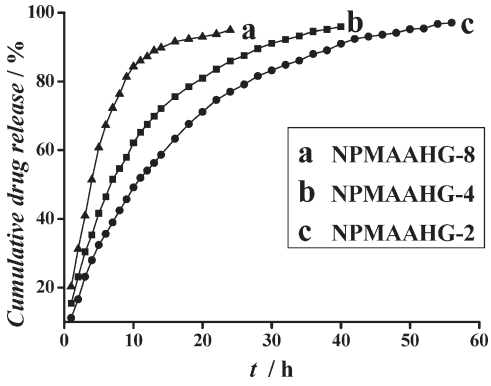

Figure 4. The in vitro drug release of IBU-loaded NPMAAHG hydrogels. a) NPMAAHG-8 in pH 7.4 buffer solution. b) NPMAAHG-4 in pH 7.4 buffer solution. c) NPMAAHG-2 in pH 7.4 buffer solution.

in SIF (pH 7.4). It takes approx. 13, 29, and 39 h to achieve 90% drug release for NPMAAHG-8, NPMAAHG-4, and NPMAAHG-2, respectively. The release rate decreases with the increasing content of the NPMA segment. That is to say, the release rate is slowed down by increasing the π–π stacking interactions in NPMAAHG. It usually takes 2–14 h to achieve 80% drug release for traditional hydrophobic-modified acrylic acid and methacrylic acid hydrogels.[21–24] Remarkably, the hydrogen bonding and π–π stacking co-switch in the network of the hydrogel successfully prolongs the release time for oral drug administration and make continuously regulated drug release possible.

2.3. Structural Characterization of NPMAAHG Hydrogels

To clearly understand the unique release behavior of the hydrogel, the structure and π–π stacking interactions and swelling behavior of the hydrogels are studied in detail. ^{1}H NMR spectra of the as-synthesized comonomer NPMA is displayed with the structure of the target molecule in Figure 5a. The chemical shift at 2.12 ppm corresponds to the protons of alkyl groups and the peaks at 5.85 and 6.34 ppm are owing to the protons on the vinyl group. Signals are found with a higher chemical shift because of the electron-withdrawing nitryl group on the phenyl ring. In addition, the peaks at 7.27 and 8.28 ppm are related to the protons on the phenyl group. FTIR spectroscopy analyses of NPMA and NPMAAHG-4 hydrogels are presented in Figure 5b. The peaks at 1530.6 and 1347.1 cm^{-1} in the bottom curve of Figure 5b correspond to the N−O asymmetrical stretching vibration of the nitryl group. The peak around 1740.7 cm^{-1} is the C=O stretching vibration, and the strong absorption bands at 1600, 864.9, and 744.5 cm^{-1} are a result of the C−H out-of-plane motion of the phenyl ring. Combining the ^{1}H NMR spectra, we can see that the functional NPMA comonomer has been successfully synthesized. The presence of O−H stretching at 3421.6 cm^{-1} in the top curve of Figure 5b clearly indicates the intercalation of MAA segments into the network of hydrogels. All the typical absorption of NPMA segments are weakened after being incorporated in PMAA hydrogels. The cleavage peaks at 1141.8 and 1102.4 cm^{-1} are attributed to the C−O stretching mode of the ester groups. The

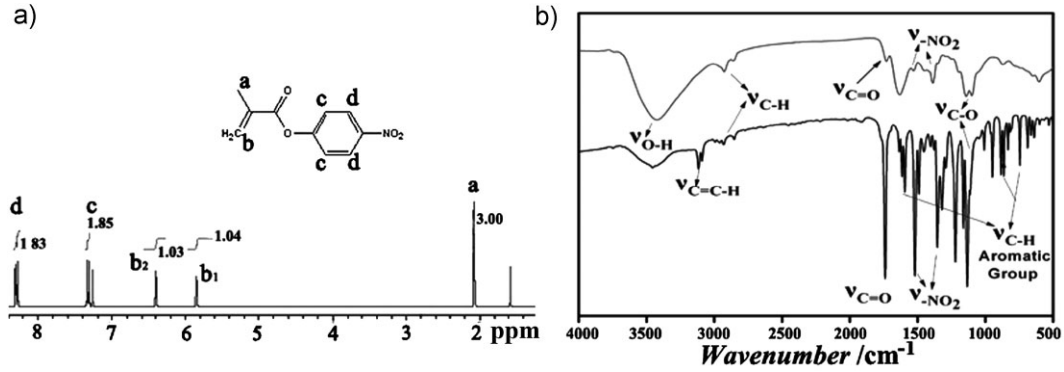

Figure 5. a) ^{1}H NMR spectra of NPMA and b) FTIR spectra of NPMA (bottom curve) and NPMAAHG-4 hydrogels (top curve). Peaks corresponding to the typical groups are singled out for further identification.

copolymerization of the monomers is revealed by the disappearance of the absorption peak at 3116.7 cm^{-1} owing to the C−H stretching vibration on unsaturated ethylenic bonds and the appearance of the C−H vibration absorption on an alkyl group around 2930 cm^{-1}. NPMA segments had been introduced in to the hydrogel system of NPMAAHG as shown by the weakened peaks at 1530.6 and 1347.1 cm^{-1} owing to the N−O asymmetrical stretching vibration of nitryl.

The lamella-like surface morphology of the as-prepared NPMAAHG-4 can be seen from the scanning electron microscopy (SEM) image (Fig. 6a). The formation of this unique surface structure may be attributable to the freeze drying process. When water is extracted from the hydrogels during the freeze drying process, the hydrogen bonding and π–π stacking interactions between the MAA and NPMA segments are formed, which lead to an ordered interconnected 3D network similar to peptide–peptide interactions as shown in Figure S1b.[31–34] X-ray diffraction (XRD) displays the periodic structure with reflections at 5.4 and 2.7 Å, which should be the separation between the planes of alkyl chains and π–π stacking groups in the NPMAAHG hydrogels in Figure 6a (insert). The extracted water forms ice layers within the hydrogels and finally evaporates, which may lead to a lamella-like morphology.[31–34] The effect of water crystallization during freezing promotes the organization of these laminate structures into oriented regularly spaced stacks, which gives rise to rather brittle freeze-dried hydrogels. Such a lamella-like structure can be formed during the freeze drying process even if there are no hydrophobic domains.[34] The presence of an absorbance peak at 1630 cm^{-1} in the FTIR spectra reveals hydrogen bonding interactions in the lamella structure (Fig. 5b).[35] The formation of π–π stacking is shown by UV-vis spectra in Figure 6b.

UV-vis spectra (Fig. 6b) reveal an absorption peak at 310 nm when the hydrogel is soaked in a pH 1.4 solution for 3 days, which is 30 nm longer than the characteristic absorption of band III in the nitrylphenyl group ($\lambda = 280$ nm). The red shift of the characteristic absorption band of the nitrylphenyl group reveals that the π–π stacking interactions between NPMA segments has been preserved after swelling in the solution of pH 1.4.[36] The characteristic absorption of band III in nitrylphenyl groups is observed at $\lambda = 280$ nm after the hydrogel is soaked in a solution of pH 7.4 for 3 days, which means that the nitrylphenyl groups are in an isolated state. In addition, the absorption peak of the hydrogel is at 295 nm after soaking in a pH 7.4 solution for 7 h, which is red-shifted by 15 nm in comparison to that of the isolated nitrylphenyl groups ($\lambda = 280$ nm), which indicates that the π–π stacking interactions between nitrylphenyl groups are partially dissociated. It is reasonable to conclude that the π–π stacking interactions help to preserve the shrinkage of the hydrogel, which are dissociated following the swelling of hydrogels because of the ionization of the cardoxylic acid groups in the network.

2.4. The Swelling Properties of the NPMAAHG Hydrogels

The swelling properties of the NPMAAHG-4 hydrogels are determined in buffer solutions from pH 1.4 to 9 with an ionic strength of 0.1 M at 37 °C (Fig. 7a). After 1 h incubation, the swelling ratios (SRs) are 1.0, 1.3, 2.9, 3.3, 4.8, 7.1, and 8.0 when exposed to solutions with pH values of 1.4, 3, 4, 5, 6, 7.4, and 9

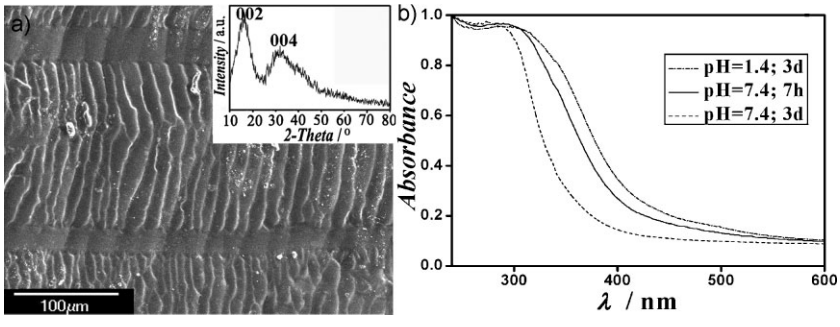

Figure 6. a) SEM images of the NPMAAHG-4 hydrogels. Insert is the XRD patterns of the synthesized NPMAAHG-4 hydrogels. b) UV-vis absorption spectra of the NPMAAHG-4 hydrogels in pH 1.4 buffer solution after 3 days incubation (dotted lines) as well as in pH 7.4 buffer solution for 7 h (solid lines) and 3 days (dashed lines).

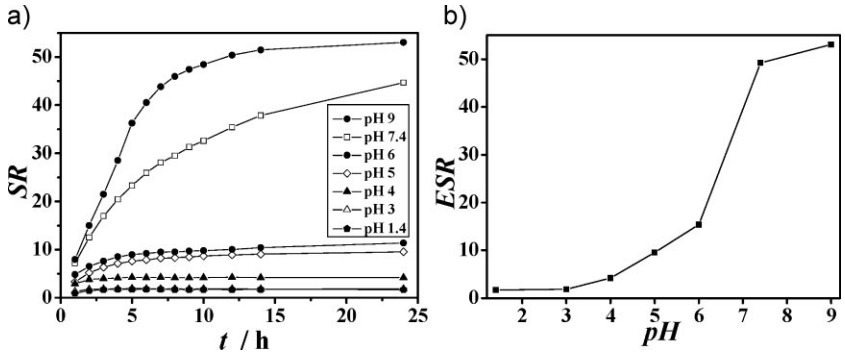

Figure 7. a) Swelling behaviors of NPMAAHG-4 in different pH buffer solutions at 37 °C, and b) ESR values of hydrogels as a function of pH.

respectively. The SR value at pH 7.4 is seven times higher than that at pH 1.4 after 1 h incubation. Notably, after 24 h incubation, the SR value (44.7) at pH 7.4 is approximately 26 times higher than the SR value at pH 1.4. Nowadays, the reported hydrophobic-modified PMAA hydrogels with a similar composition exhibit an SR value of 1.0–3.0 at pH 1.4 and 3.0–25 at pH 7.4.[21–24] We can see that the equilibrium swelling ratios (ESRs) of the NPMAAHG-4 hydrogels increase dramatically when the pH value is above the pK_a of the carboxylic acid (pK_a = 5.5) (Fig. 7b).[11] The time taken for the NPMAAHG-4 hydrogels to reach the equilibrium swelling is 74, 39, and 24 h at pH 6, 7.4, and 9, respectively. The forgoing results show that the pH response sensitivity of the NPMAAHG-4 hydrogel is greatly improved after introducing π–π stacking interactions. NPMA segments in the network of the NPMAAHG hydrogels form strong π–π stacking domains,[9,10,23] which are gradually weakened as the hydrogels swell.

2.5. Kinetic Analysis of the Co-switched Drug Release Systems

A kinetic analysis has been carried out of the in vitro drug release curves of IBU-loaded NPMAAHG-*n* hydrogels and the resulting kinetic parameters are listed in Table 1. The general solute release behaviors of the polymeric hydrogels are analyzed by fitting the Peppas equation:[37,38]

$$M_t/M_\infty = kt^n \tag{1}$$

where n is a diffusion exponent that determines the dependence of the release rate on time, k is a release rate constant, and M_t and M_∞ correspond to the cumulative mass of IBU released at a given time t and the cumulative release at time infinity, respectively.

To illustrate the influence of the swelling behavior on the solute release in modified PMAA-based hydrogels, the solute release mechanisms are analyzed by Equation (2):[39]

$$S_W = \frac{\nu \cdot \delta(t)}{D_s} = \frac{\text{Solvent motion}}{\text{Soluted diffusion}} \tag{2}$$

where the swelling interface number S_w is a dimensionless number that provides the relative velocity between solvent and solute, ν represents the velocity of the swelling interface, δ is the thickness of the swollen layer, and D_s is the drug diffusion coefficient in the copolymer that can be determined experimentally by using diffusion Equation (3):[29,38]

$$D_s = \frac{\pi L^2}{16}\left[\frac{d}{dt^{1/2}}\left(\frac{M_t}{M_\infty}\right)\right]^2 \tag{3}$$

where L is the thickness of the sample. Equation (4) is employed to calculate the velocity of the swelling interface (ν):

$$\nu = \left(\frac{dW_s}{dt}\right)/(2A\rho_s) \tag{4}$$

where (dW_s/dt) is the weight of water absorbed by the hydrogels per unit time, ρ_s is the density of water at 37 °C, A is the area of one face of the slab, and the factor 2 accounts for the fact that the diffusion takes place through both faces.

The thickness of the swollen layer $\delta(t)$ is a variable parameter that changes with time and reaches a maximum value δ_{max}, which is correlated with the swelling ratio (SR):

$$\frac{\delta_{max}}{L} = \left[1 + \left(\frac{\rho_p}{\rho_s}\right)\text{ESR}\right] \tag{5}$$

where ρ_p and ρ_s are the densities of dry polymer and penetrant, and ESR is the equilibrium swelling ratio value.

The release kinetic parameters of the NPMAAHG-4 release system in solutions at pH 1.4 and 7.4 are compared in Table 1. The diffusion exponent (n) is 0.492 in pH 7.4 solutions, which means a

Table 1. Release nature of IBU from the NPMAAHG hydrogels.

Sample	pH [a]	n [b]	$D_s \times 10^{-7}$[c] [cm^2 min^{-1}]	$\nu \times 10^{-5}$ [d] [cm min^{-1}]	δ_{max} [e] [cm]	S_w [f]
NPMAAHG-4	SGF 1.4	0.345	0.27	1.47	0.129	69.1
NPMAAHG-2	SIF 7.4	0.398	5.77	5.53	0.311	29.8
NPMAAHG-4	SIF 7.4	0.492	13.2	6.85	0.318	16.5
NPMAAHG-8	SIF 7.4	0.546	37.4	8.92	0.326	7.8
PMAAHG	SIF 7.4	0.691	39.1	8.48	0.303	5.95

[a] pH refers to the pH values of the release medium. [b] n is the diffusion exponent calculated from Equation (3). [c] D_s is the drug diffusion coefficient in the polymer. [d] ν stands for the velocity of the swelling interface. [e] δ_{max} represents the equilibrium thickness of the swollen layer. [f] S_w is the swelling interface number.

typical Fickian diffusion behavior; while the diffusion exponent is 0.345 in pH 1.4 solutions, which means that the Fickian diffusion is smaller than that in pH 7.4 solutions. The results indicate that the pore size of the swelled gel network at pH 7.4 is just large enough to transfer guest molecules through and it is not restricted by the relaxation of the polymer.[40] The pore size of the swollen gel network at pH 1.4 should be smaller than that at pH 7.4, which can be seen from the different equilibrium thickness of the swollen layers (δ_{max} is 0.129 cm in pH 1.4 solution, δ_{max} is 0.318 cm in pH 7.4 solution) (Table 1). The depressed swelling in the pH 1.4 solution leads to the small pore size of the gel network and thus dramatically decreases the drug diffusion coefficient ($D_s = 2.76 \times 10^{-8}$ cm^2 min^{-1} in pH 1.4 solution, $D_s = 1.32 \times 10^{-6}$ cm^2 min^{-1} in a pH 7.4 solution). The velocity of the swelling interface (v) at pH 1.4 (1.474×10^{-5} cm min^{-1}) is slower than that at pH 7.4 (6.850×10^{-5} cm min^{-1}) as a result of the non-ionization of MAA units in low pH solutions, which may lower the osmotic pressure in and out of the hydrogels and decrease the mobility of water molecules in the hydrogels.[41] More interestingly, the swelling interface number (S_w) in a pH 1.4 solution (69.14) is much higher than that in a pH 7.4 solution (16.52), which reveals that the pH-inactive π–π stacking co-switched hydrogels are very effective in holding back the release of the drugs at lower pH even if they have a relatively weak influence on the penetration of water.

To investigate the continuous regulation behavior of the hydrogen bonding and π–π stacking switches on the release of drugs, the release kinetics of copolymers with different NPMA and MAA molar ratios are investigated (Table 1). With the increase of NPMA groups in the NPMAAHG-*n* co-polymers, the release behavior of the copolymers changes from non-Fickian diffusion to Fickian diffusion as revealed by the decreasing diffusion exponents (n) of NPMAAHG-8, NPMAAHG-4, and NPMAAHG-2 from 0.546, to 0.492, and 0.398, and the velocity of the swelling interface (v) decreases from 8.922×10^{-5}, 6.850×10^{-5}, to 5.525×10^{-5} cm min^{-1}. The equilibrium thickness of the swollen layer (δ_{max}) decreases with the increase of NPMA groups, which means that a little swelling and a rearrangement of the gels is required to accommodate the solvent for NPMAAHG-*n* with less MAA segments.[42,43] Along with the decrease in swelling ratios and swelling rates of hydrogels, the drug diffusion coefficient (D_s) reduces dramatically from 3.74×10^{-6}, 1.32×10^{-6}, to 5.77×10^{-7} cm^2 min^{-1}. The dramatic changes in D_s cause the swelling interface number S_w to decrease from 29.82, 16.52, to 7.78, which further reflects a diffusion kinetics conversion from non-Fickian to Fickian diffusion.[37,38] Hydrogen bonds and π–π stacking are competing factors to control the release behavior of the hydrogel. The latter is of a hydrophobic nature and leads to stacking in aqueous solution to hold back the release of drugs; whilst the former is of a hydrophilic nature and leads to water being imbibed which induces the swelling of the hydrogels. The π–π stacking helps to sustain the structure of the hydrogels in water, thus the drug release occurs in the style of Fickian diffusion when the π–π stacking predominates in the polymers. When hydrogen bonding predominates in the hydrogels, water will penetrate into the hydrogels and results in the swelling process which boosts the release of drugs locked in the polymer, thus the drug release occurs in the style of non-Fickian diffusion. Therefore, by concocting polymers with hydrogen bonding switches and π–π stacking switches, we can achieve a continuous control over the delivery of guest molecules and make it more suitable for systematic administration, as shown in the present work.

To distinguish the influence of EDGMA crosslinking and that of hydrophobic π–π stacking domains on the sustained release behavior, PMAAHG hydrogels without π–π stacking interactions have been synthesized with more EDGMA crosslinkers (1.05 wt %) than that of NPMAAHG-*n* (1.00 wt%). As was expected, the swelling of the PMAAHG hydrogels with more EDGMA crosslinkers is depressed, and display a decreased equilibrium thickness of the swollen layer ($\delta_{max} = 0.303$ cm) compared with NPMAAHG-*n* (Table 1). The increased number of crosslinker units weaken the swelling of the hydrogel as revealed by the slow velocity of the swelling interface ($v = 8.48 \times 10^{-5}$ cm min^{-1}). However, PMAAHG hydrogels display non-Fickian diffusion with a diffusion exponent of 0.691, which is higher than that of NPMAAHG-*n*. The increased diffusion exponent ($n = 0.691$) indicates an increased release of drugs as shown in Figure S2 (see supporting information). Moreover, the drug diffusion coefficient (D_s) of PMAAHG is 39.1, which is higher than that of NPMAAHG-*n*. The foregoing results show that an increase in crosslinking may be effective to decrease the swelling but may not be effective to sustain drugs. That is to say, the drug retention in the NPMAAHG-*n* is related to the presence of hydrophobic π–π stacking domains rather than simple differences in swelling.

3. Conclusion

The drug release can be continuously controlled by incorporating hydrogen bonding and π–π stacking interactions together in NPMAAHG hydrogels. The properties of the NPMAAHG hydrogels can be dramatically tailored by varying the ratio of hydrogen bonding and π–π stacking interactions, which exhibit high pH sensitivity, and excellent swelling and desired drug release behavior. The drug delivery behavior can be regulated to correspond with the transit time of the gastrointestinal tract. The improvement in efficiency of controlling drug release is attributed to the competing effect between hydrogen bond switches and π–π stacking switches, which changes the release kinetics from non-Fickian diffusion to Fickian diffusion. This present work provides a pathway to continuously regulate hydrogels for prolonged sustained drug release in oral chemotherapy. This co-switched hydrogel should be an effective sustained drug release system particularly well-suited for systematic administration.

4. Experimental

Materials: EGDMA (98%) was purchased from J&K Chemical Company (J&K Chemical Ltd). Methacryloyl chloride (C_4H_5ClO, CP) was received from Shanghai Haiqu Chemical Co., Ltd. Other regents such as triethanolamine ($N(OC_2H_5)_3$, AR), 4-nitrophenol ($C_6H_5NO_3$, AR), α-methacrylic acid ($CH_2{=}C(CH_3)COOH$, CP), dichloromethane (AR), *N*,*N*-dimethylformamide (DMF, AR), 2,2′-azoisobutyronitrile (AIBN, AR), and ibuprofen ($C_{13}H_{18}O_2$, AR) were supplied by Sinopharm Chemical Reagent Company (Sinopharm Chemical Reagent Co., Ltd). They were all directly used without further purification.

Synthesis of Functionalized Comonomer NPMA: The functionalized monomer NPMA was fabricated by adding $N(OC_2H_5)_3$ to a mixture of

methacryloyl chloride, 4-nitrophenol, and dichloromethane (40 mL). The reaction was performed in an ice-water bath for about 24 h with a feed composition at a 1:1:1 mol ratio. Saturated NH_4Cl solution (40 mL) was used to terminate the reaction. Substrates were extracted by an appropriate amount of CH_2Cl_2, and then recrystallized from ethanol to obtain the final functionalized monomer. The product was characterized by ^{1}H NMR spectroscopy and FTIR spectroscopy to certify its structure.

Preparation of the NPMA-Modified Copolymer Hydrogel NPMAAHG: In order to prepare NPMA-modified copolymer hydrogels, as-synthesized NPMA was reacted with MAA by using EGDMA as a crosslinker. In detail, a calculated amount of NPMA and α-methacrylic acid with a molar ratio of 1:4 were first dissolved in 1 mL of DMF. Afterwards, EGDMA (1 wt%) was added to the above solution with a microsyringe. The mixture was bubbled with dried nitrogen gas for 20 min to remove oxygen. Afterward, the oil-soluble initiator AIBN was adding, and the resultant solution was bubbled for another 20 min with dried nitrogen gas. The polymerization was performed in a glass test tube (20 mm diameter and 180 mm length). The polymers were removed from the mold after reaction and soaked in a water/acetone (50:50 v/v) solvent mixture to extract the unreacted compound and the solvent was replaced daily for at least one week until there were no detectable substances. After extraction, the hydrogels (NPMAAHG) were cut into slabs and dried in oven at 40 °C until there was no detectable weight loss (with a thickness between 0.081 and 0.110 cm). PMAA hydrogels (PMAAHG) without NPMA segments were synthesized under the same conditions as those for NPMAAHG. SEM, UV-Vis spectroscopy, XRD, and FTIR spectroscopy were used to investigate the structure of the as-synthesized hydrogels. Samples for UV-vis spectroscopy were immersed in the relevant solutions for 3 days before the examination.

Swelling Property Experiments of NPMAAHG Hydrogels: The dried slabs were incubated in buffer solutions with different pH at 37 °C. They were carefully taken out from the solution when a desired incubation time was reached, wiped gently with a filter paper to remove of free liquid on the surface and then weighed. The degree of swelling was calculated by measuring the weight of the swollen plates until there was no detectable weight change. The following equation was used:

$$\text{Swelling ratio} = \frac{W_s - W_d}{W_d} \tag{6}$$

where W_s is the weight of the swollen gels and W_d represents the weight of the dried gels. When a hydrogel reaches its swelling equilibrium state under a fixed condition, its swelling ratio is called the ESR. In general, equilibrium was reached within 3 days. All measurements were performed in triplicate for each sample.

Preparation of Buffer Solutions: KCl/HCl, pH 1.4; $HCl/KHC_6H_4(COO)_2$/NaOH, pH 3.0–5.0; $NaOH/KH_2PO_4/NaHPO_4$, pH 6.0–7.4; $NaBO_4$/NaOH, pH 9. For pH sensitivity investigations, the ionic strength of the buffer solution was adjusted to a constant value of 0.1. For swelling experiments in a salt solution, KCl was introduced into the solutions to achieve the required ionic strength.

Drug Loading and Release Experiments: For drug release experiments, IBU was used as a model drug. The dried NPMAAHG hydrogels were equilibrated in 20 mL of acetone with ibuprofen (0.8 g) for 3 days. The drug-loaded hydrogels were air dried in an oven at 40 °C for 48 h, afterwards they were weighed and the IBU loading ratio of the system was calculated.

Release experiments for IBU were carried out in bottles filled with 40 mL of SIF (pH 7.4) and SGF (pH 1.4) at 37 °C. An aliquot volume was withdrawn from every release medium at a given time and at the same time an equal amount of fresh solution was added. As a control experiment, drug-loaded PMAA hydrogels with different degrees of crosslinking were carried out under the same conditions as for the NPMAAHG hydrogels.

Release experiments of IBU from the NPMAAHG-4 hydrogel system by simulating the gastrointestinal tract were started with immersion in a solution at pH 1.4. After 3 h, the solution was adjusted to a high pH value to open the pathways. The drug release amount was measured by UV-vis spectroscopy at about 264 nm. The cumulative fraction of the IBU was calculated by the following equation:

$$\text{Cumulative drug release} = M_t/M_\infty \tag{7}$$

where M_t and M_∞ corresponds to the cumulative mass of IBU released at time t and the cumulative release at time infinity, respectively.

Characterization: SEM was performed using a JXA-8100, operated at an accelerating voltage of 20 kV. Samples were freeze-dried for 24 h before being coated with 10 nm of Au to prevent charging. XRD patterns were recorded on a D/max 2550V diffractometer with Cu Kα radiation ($\lambda = 1.542$ Å). ^{1}H NMR spectroscopy was carried out on Varian Mercury vx at 300 MHz. Samples were dissolved in deuterochloroform ($CDCl_3$) to remove the influence of the solution. FTIR analysis was carried out using KBr discs in the region of 4000–400 cm^{-1} by a SHIMADZU (IR Prestige-21). The pH value was measured on a Model PHS-25 pH Meter Instruction (with the precision of 0.1). UV-vis spectroscopy was performed on a U-4100 spectrometer using a pair of quartz cuvettes ($12.4 \times 12.4 \times 45$ mm^3).

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