

Preparation and Drug-Release Behavior of Minocycline-Loaded Poly[hydroxyethyl methacrylate-co-poly(ethylene glycol)-methacrylate] Films

Gülay Bayramoğlu,¹ Ertan Batislam,² M. Yakup Arica¹

¹Biochemical Processing and Biomaterial Research Laboratory, Faculty of Arts and Sciences, Gazi University, 06500 Teknik Okullar, Ankara, Turkey

²Department of Urology, Faculty of Medicine, Kırıkkale University, 71100 Kırıkkale, Turkey

Received 24 February 2008; accepted 15 October 2008

DOI 10.1002/app.29450

Published online 23 January 2009 in Wiley InterScience (www.interscience.wiley.com).

ABSTRACT: In this work, biocompatible hydrogel matrices for wound-dressing materials and controlled drug-release systems were prepared from poly[hydroxyethyl methacrylate-co-poly(ethylene glycol)-methacrylate] [p(HEMA-co-PEG-MA)] films via UV-initiated photopolymerization. The characterization of the hydrogels was conducted with swelling experiments, Fourier transform infrared spectroscopy, scanning electron microscopy, thermogravimetric analysis (differential scanning calorimetry), and contact-angle studies. The water absorbency of the hydrogel films significantly changed with the change of the medium pH from 4.0 to 7.4. The thermal stability of the copolymer was lowered by an increase in the ratio of poly(ethylene glycol) (PEG) to methacrylate (MA) in the film structure. Contact-angle measurements on the surface of the p(HEMA-co-PEG-MA) films demonstrated that the

copolymer gave rise to a significant hydrophilic surface in comparison with the homopolymer of 2-hydroxyethyl methacrylate (HEMA). The blood protein adsorption was significantly reduced on the surface of the copolymer hydrogels in comparison with the control homopolymer of HEMA. Model antibiotic (i.e., minocycline) release experiments were performed in physiological buffer saline solutions with a continuous flow release system. The amount of minocycline release was shown to be dependent on the HEMA/PEG-MA ratio. The hydrogels have good antifouling properties and therefore are suitable candidates for wound dressing and other tissue engineering applications. © 2009 Wiley Periodicals, Inc. *J Appl Polym Sci* 112: 1012–1020, 2009

Key words: biocompatibility; biomaterials; drug delivery systems; films; hydrophilic polymers

INTRODUCTION

Hydrogels are polymeric materials that do not dissolve in water but swell considerably in aqueous media and show a minimal tendency to adsorb proteins from bodily fluids because of their low interfacial tension.^{1–6} The structural features of these materials dominate their surface properties, permselectivity, and permeability, giving hydrogels their unique, interesting properties and the similarity of their physical properties to those of living tissue.⁷ Because of their high water contents, low water contact angles, high permeability, and low friction coefficients, hydrogels are studied extensively as replacements for soft tissue. Hydrogels are of special interest in controlled drug-release applications because of their soft tissue biocompatibility, the ease with which the drugs are dispersed in the matrix, and the high degree of control achieved by the selec-

tion of the physical and chemical properties of the polymer network.^{8,9}

The homopolymer and copolymer hydrogels of 2-hydroxyethyl methacrylate (HEMA) have found extensive applications in the field of biomedicine because of their good chemical stability and high biocompatibility.^{10,11} Poly(ethylene glycol) (PEG) is a water-soluble, nontoxic, and nonimmunogenic polymer. Surfaces containing PEG are interesting biomaterials because they exhibit low degrees of protein adsorption and cell adhesion.^{12,13} Because of its biocompatibility and safe toxicity profile, it is applied in various biomedical areas such as drug delivery,¹⁴ wound healing, and tissue repair systems.^{15–20}

In recent years, there has been increased interest in the controlled release of drugs, which is another efficient technique for the use of medicines. The ideal drug delivery system should be inert, biocompatible, mechanically strong, comfortable for the patient, capable of achieving a high drug loading for the required blood levels, immune to accidental release, simple to apply, and easy to fabricate.^{1,2}

Minocycline is a synthetic tetracycline-derived antibiotic. The tetracyclines, including minocycline,

Correspondence to: G. Bayramoğlu (g_bayramoglu@hotmail.com).

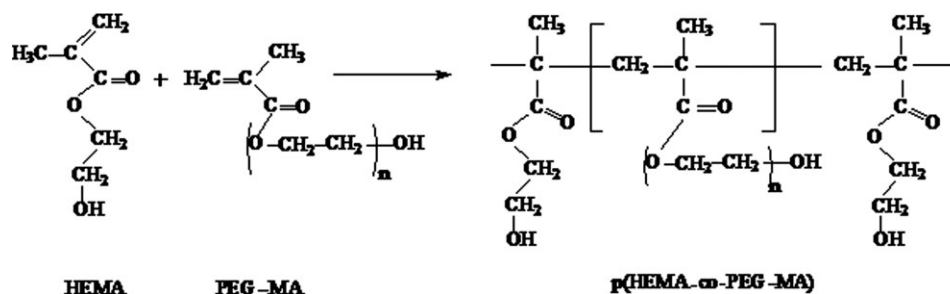


Figure 1 Chemical structure of the p(HEMA-*co*-PEG-MA) film.

have similar spectra of antimicrobial activity against a wide range of gram-positive and gram-negative organisms. They eliminate bacteria that cause pneumonia, infections of the skin, and genitourinary and central nervous system infections.¹⁶

In this study, hydrogel networks consisting of poly(hydroxyethyl methacrylate) (pHEMA) and poly(ethylene glycol)-methacrylate (PEG-MA) in various proportions were prepared by UV-initiated photopolymerization for the construction of a controlled drug-release system. The hydrogel networks were characterized with differential scanning calorimetry (DSC), scanning electron microscopy, and contact-angle studies. We also studied some factors in a continuous drug-release system that may have an influence on drug release from a poly[hydroxyethyl methacrylate-*co*-poly(ethylene glycol)-methacrylate] [p(HEMA-*co*-PEG-MA)] hydrogel network, such as the ratio of HEMA to PEG-MA and the loaded amount of the model antibiotic (i.e., minocycline). Finally, the kinetics of antibiotic release from p(HEMA-*co*-PEG-MA) were analyzed.

EXPERIMENTAL

Materials

HEMA, PEG-MA, and α - α' -azobisisobutyronitrile were obtained from Sigma-Aldrich Chemicals GmbH (Erkerode, Germany). HEMA monomer was purified by distillation under reduced pressure. Minocycline hydrochloride was supplied by Sigma-Aldrich and used as received. All other chemicals were analytical-grade and were purchased from Merck AG (Darmstadt, Germany). The water used in this work was purified with a Barnstead (Dubuque, IA) ROPure LP reverse osmosis system.

Synthesis of the p(HEMA-*co*-PEG-MA) drug-release system

The pHEMA and p(HEMA-*co*-PEG-MA) films were prepared by UV-initiated photopolymerization. The chemical structure of the copolymer film is presented in Figure 1. The polymerization was carried out in a flat glass mold (internal diameter = 9 cm).

To check the effect of the monomer ratio on the film properties and drug-release efficiency in the initial polymerization mixture, three different HEMA/PEG-MA ratios were used (1, 2.0 : 1.0 v/v; 2, 1.5 : 1.5 v/v; and 3, 1.0 : 2.0 v/v). The polymerization mixture was purged with nitrogen for 5 min and transferred into molds. After sealing, it was exposed to UV radiation (TUW series, Philips; UV-C, 254-nm short wave, 24 W, Ankara, Turkey) for 30 min. For the preparation of drug-loaded films of p(HEMA-*co*-PEG-MA)-1 to p(HEMA-*co*-PEG-MA)-3, 5–20 mg/mL minocycline (Fig. 2) was added to the polymerization mixture. Polymerization was carried out as described previously.

Characterization studies of the hydrogel films

The density of the hydrogel films was determined with a Gay-Lussac pycnometer (Dahlewitz, Germany), and *n*-decane was chosen as the inert liquid, which is a nonsolvent for the hydrogel. The thickness of the p(HEMA-*co*-PEG-MA) film was estimated with a micrometer thickness gauge. The morphology of pHEMA and its drug-loaded counterpart was investigated with a JEOL (Tokyo Japan) model JSM 5600 scanning electron microscope. The dried polymer films were mounted on the base plate and then coated with gold *in vacuo*. The morphology and surface structure of the films were obtained at the required magnification at room temperature.

Fourier transform infrared (FTIR) spectra of the pHEMA and p(HEMA-*co*-PEG-MA)-3 films were obtained with an FTIR spectrophotometer (FTIR 8000 series, Shimadzu, Tokyo, Japan). The dry sample (ca. 0.01 g) was mixed with KBr (0.1 g) and pressed into

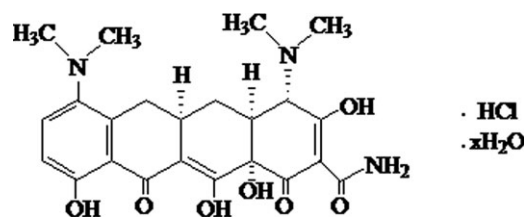


Figure 2 Chemical structure of minocycline.

a tablet form. The FTIR spectrum was then recorded in the wavelength region of 400–4000 cm^{-1} .

The thermal analysis was performed with a Pyris Sapphire differential scanning calorimeter (Standard 115V, Yokohawa, Japan) at a heating rate of 10°C/min under a nitrogen atmosphere. Samples were embedded in nonhermetic aluminum pans.

The water content of the p(HEMA-*co*-PEG-MA) films was determined at room temperature in different buffer solutions (50 mM, pH 4.0–7.4) with a gravimetric method. The preweighed dry samples were immersed in buffer solutions; the swollen films were removed after the excess surface-adhered liquid dropped, and they were weighed on a sensitive balance ($\pm 1.0 \times 10^{-4}$ g; model AX 120, Shimadzu). To ensure complete equilibration, the film samples were allowed to swell for 48 h.

Determination of the contact angles of the films

The contact angles of the materials to different test liquids (i.e., water, glycerol, and diiodomethane) were measured by the sessile drop method at 25°C with a CAM 200 digital optical contact-angle meter (KSV Instruments, Ltd., Helsinki, Finland). Both the left and right contact angles and drop dimension parameters were automatically calculated from the digitalized image. The measurements were the averages of at least five contact angles taken on three membrane samples.

Determination of the antifouling properties of the films

To determine the antifouling properties of the films of poly(HEMA) and p(HEMA-*co*-PEG-MA)-1 to p(HEMA-*co*-PEG-MA)-3, the adsorption of human serum albumin (HSA) and fibrinogen was studied at 37°C for 18 h in a batch system. The initial concentration of protein was 0.5 mg/mL in the individual adsorption medium. The amount of adsorbed protein on the film surface was obtained with the following equation:

$$q = [(C_0 - C)V]/S \quad (1)$$

where q is the amount of protein adsorbed onto the film surface ($\mu\text{g}/\text{cm}^2$); C_0 and C are the concentrations of protein in the solution before and after adsorption, respectively ($\mu\text{g}/\text{mL}$); V is the volume of the protein solution; and S is the surface area of the film. The amount of protein adsorbed onto the films was determined with a fluorescence spectrofluorometer (FP-750, Jasco, Tokyo, Japan). The fluorescence intensity was measured with excitation and emission at 280 and 340 nm, respectively. Calibration curves were prepared from HSA and fibrinogen and were used in the calculation of the protein content of the solutions.

Controlled release studies of minocycline from the hydrogel films

The *in vitro* release studies were conducted in a continuous flow system with constant stirring. Minocycline-loaded film discs of p(HEMA-*co*-PEG-MA)-1 to p(HEMA-*co*-PEG-MA)-3 (2.0 g) were placed in the continuous flow drug-release cell, and a physiological buffer solution (pH 7.4) was introduced through the bottom inlet port of the flow cell at a flow rate of 10 mL/h at 37°C with a peristaltic pump (model IPC, Ismatec, Düsseldorf, Germany). The effluent was collected at 1.0-h intervals, and the released minocycline (at a maximum wavelength of 266 nm) was determined spectrophotometrically (model 1601, Shimadzu). The analyzed solution was added back to the collected dissolution media to maintain a constant volume. To study the effect of the minocycline loading on the release rate, similar procedures were followed for 5, 10, 15, and 20 mg/mL drug-loaded samples. All experiments were repeated four times to minimize the variational error. Therefore, the data presented in the graphs show the average values of four experiments.

The model drug concentration in the collected dissolution medium was calculated with a calibration curve obtained from samples of known concentrations, and the controlled release profiles were normalized to 100% drug release at the end of the experiment (time $t = \infty$).

The percentage of the drug-loading efficiency in the hydrogels was then calculated as follows:

$$\begin{aligned} \text{Loading efficiency}(\%) \\ = \text{Practical drug loading} / \text{Theoretical drug loading} \\ \times 100 \quad (2) \end{aligned}$$

RESULTS AND DISCUSSION

Properties of the p(HEMA-*co*-PEG-MA) hydrogels

PEG-carrying films of p(HEMA-*co*-PEG-MA)-1 to p(HEMA-*co*-PEG-MA)-3 were prepared with UV-initiated photopolymerization. The hydrogel film thickness and density were measured to be 600 μm and 1.017 g/cm^3 , respectively. The equilibrium swelling of p(HEMA-*co*-PEG-MA) was dependent on the PEG-MA ratio in the copolymer structure and medium pH (Table I). As expected, the introduction of PEG-MA into the HEMA hydrogel system altered the bulk properties of the resulting gels. The PEG chains of PEG-MA were expected to be spread over the gel surface because the pendant PEG components were more hydrophilic than the copolymer backbone, as evidenced by the swelling behavior and contact-angle data. As seen in Table I, the degree of swelling of the films of p(HEMA-*co*-PEG-

TABLE I
Equilibrium Water Content for the Films of pHEMA and p(HEMA-co-PEG-MA)-1 to p(HEMA-co-PEG-MA)-3 at Different pHs

Film sample	Equilibrium water content (%)				
	pH 4.0	pH 5.0	pH 6.0	pH 7.0	pH 7.4
pHEMA	54.2 ± 1.4	55.9 ± 2.3	57.4 ± 1.7	60.8 ± 1.1	64.3 ± 2.1
p(HEMA-co-PEG-MA)-1	69.7 ± 2.4	74.0 ± 2.1	75.6 ± 1.1	77.6 ± 1.3	81.0 ± 1.7
p(HEMA-co-PEG-MA)-2	79.2 ± 1.8	82.4 ± 2.2	88.0 ± 2.1	93.5 ± 2.1	98.4 ± 1.1
p(HEMA-co-PEG-MA)-3	108.4 ± 3.1	115.7 ± 3.1	124.4 ± 2.3	135.2 ± 1.4	137.1 ± 2.3

DIM, diiodamethane; γ_1 , surface tension of test liquid.

MA)-1 to p(HEMA-co-PEG-MA)-3 were determined to be between 81.0 ± 1.7 and $137.1 \pm 2.3\%$ at pH 7.4, and the equilibrium swelling values were found to be lower in an acidic solution (pH = 4.0–6.0) than in a neutral pH or slightly alkaline pH because the equilibrium swelling value for pHEMA (at pH 7.4, $64.3 \pm 2.1\%$) was lower than those for the copolymers. As is known, pHEMA is practically not pH-sensitive, whereas the equilibrium water contents of pHEMA and p(HEMA-co-PEG-MA) exhibited a small increase with increasing pH (Table I). Similar observations have been reported in previous studies.^{21,22}

Figure 3(A,B) shows scanning electron micrographs of the p(HEMA-co-PEG-MA)-3 hydrogel and minocycline-loaded counterpart, respectively. Scan-

ning electron micrographs showed a slight difference in the surface morphology of the films versus the drug-loaded counterparts. The hydrogel surface was rough and nonporous. A uniform distribution of the drug in the film was also observed.

The FTIR spectra of both pHEMA and p(HEMA-co-PEG-MA)-3 had the characteristic stretching vibration band of hydrogen-bonded alcohol (O–H) around 3550 cm^{-1} , and the C=O stretching vibration of the ester group also appeared at 1720 cm^{-1} (data not shown). On the other hand, several bands appeared in the fingerprint region for PEG between 1600 and 1200 cm^{-1} on the p(HEMA-co-PEG-MA)-3 structures. These peaks were assigned to the $-\text{CH}_2$ scissoring band of PEG at 1450 cm^{-1} and the anti-symmetric and symmetric stretching bands ($-\text{OCH}_2-\text{CH}_2$) of PEG at 1360 and 1275 cm^{-1} , respectively. It should be noted that the hydrogen-bonded alcohol (O–H) stretching band intensity of p(HEMA-co-PEG-MA)-3 films was higher than that of pure pHEMA because of the incorporation of PEG-MA into the copolymer structure.

The thermal stability of the pure pHEMA and p(HEMA-co-PEG-MA)-3 copolymer was evaluated with thermogravimetric analysis. The DSC curves of pure pHEMA and p(HEMA-co-PEG-MA)-3 films are shown in Figure 4. pHEMA shows a distinct feature in the DSC curve, which has one endotherm at about 338°C due to the thermal degradation and another

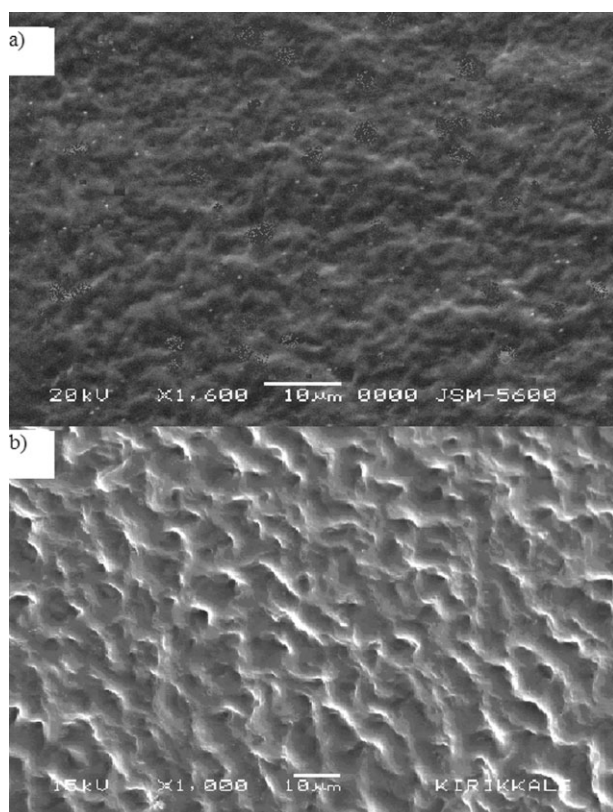


Figure 3 Scanning electron micrographs of the (a) control p(HEMA-co-PEG-MA)-3 and (b) minocycline-loaded p(HEMA-co-PEG-MA)-3.

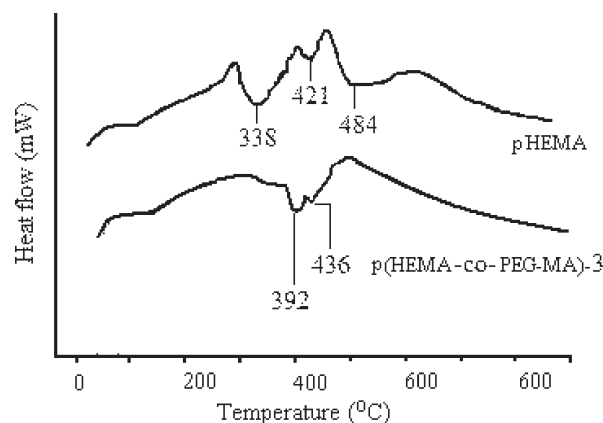


Figure 4 Results of the DSC measurements: p(HEMA) and p(HEMA-co-PEG-MA)-3 films.

TABLE II
Contact Angles of the Test Liquids on the Films of pHEMA and p(HEMA-co-PEG-MA)-1 to p(HEMA-co-PEG-MA)-3

Film sample	Test liquid and surface tension		
	Water ($\gamma_1 = 71.3$)	Glycerol ($\gamma_1 = 64.0$)	DIM ($\gamma_1 = 50.8$)
pHEMA	57.5 \pm 0.3	54.6 \pm 0.3	35.3 \pm 0.4
p(HEMA-co-PEG-MA)-1	49.8 \pm 1.4	63.7 \pm 1.8	36.9 \pm 1.1
p(HEMA-co-PEG-MA)-2	46.5 \pm 1.6	65.8 \pm 0.9	39.6 \pm 0.9
p(HEMA-co-PEG-MA)-3	43.7 \pm 0.9	66.1 \pm 0.5	41.4 \pm 1.3

DIM, diiodomethane; γ_1 , surface tension of test liquid.

endotherm peak around 484°C. In the case of p(HEMA-co-PEG-MA)-3, four endothermic peaks can be observed. The thermal degradation peak decreased about 48°C, and the decomposition temperature was lowered to about 436°C. A similar observation was reported in our previous study.^{23,24}

The results for the contact-angle measurements of water, glycerol, and diiodomethane on film surfaces of pHEMA and p(HEMA-co-PEG-MA)-1 to p(HEMA-co-PEG-MA)-3 are presented in Table II. All PEG-MA-carrying materials showed lower water contact angles than the pure pHEMA. All the investigated samples yielded a different contact angle. As expected, pHEMA had a polar surface and showed a water contact angle of 57.5°. As can be seen in Table II, for the polymer series of p(HEMA-co-PEG-MA)-1 to p(HEMA-co-PEG-MA)-3, the contact angles of the three liquids depended on the PEG-MA monomer ratio of the materials. As the PEG-MA ratio increased (2 : 1, 1.5 : 1.5, and 1 : 2 v/v HEMA/PEG-MA) in the copolymer structure, the water contact angles decreased (Table II). Contact-angle measurements are used in the characterization of material surfaces to describe hydrophilicity or to estimate the surface free energy. The wettability of biomaterial surfaces can be examined by a comparison of the contact angles for water and diiodomethane because these two solvents are often used as reference liquids in analyses of interactions of polar and apolar solvents with solid surfaces. As presented in Table II, all the PEG-carrying, comonomer-incorporated hydrogel materials showed higher glycerol contact angles than pHEMA. This could be due to the increased density of polar PEG macromole-

cules on the copolymer surfaces as the PEG-MA ratio increased.

Protein adsorption studies

The antifouling properties of p(HEMA-co-PEG-MA)-1 to p(HEMA-co-PEG-MA)-3 were tested with two important plasma proteins (i.e., serum albumin and fibrinogen) in a batchwise manner with pure pHEMA as a control system. Drug-loaded samples were incubated with a physiological buffer solution (pH 7.4) containing a 0.5 mg/mL concentration of an individual plasma protein for a period of 18 h at 37°C. The two tested proteins reached their maximum adsorption values on pHEMA and p(HEMA-co-PEG-MA)-1 to p(HEMA-co-PEG-MA)-3 after 10 h and remained constant after this period. The amounts of adsorbed serum albumin and fibrinogen on the sample surface are presented in Table III.

The isoelectric points of the albumin and fibrinogen were 4.0 and 6.0, respectively, so these proteins carried a negative charge at a blood pH of 7.4. The relatively high basic [Lewis base electron donor (γ^-)] component of the PEG-carrying films could create a repulsive force against the negatively charged blood proteins. Thus, an increase in the PEG-MA monomer ratio could improve the antifouling properties of the materials. As shown in Table III, the amounts of albumin and fibrinogen adsorbed onto p(HEMA-co-PEG-MA)-1 to p(HEMA-co-PEG-MA)-3 were reduced versus pure pHEMA. The reduction in the adsorbed amount of proteins was mainly due to the repulsion of the Lewis base component of the PEG molecules.¹⁷ In this work, the hydrophilic PEG

TABLE III
Drug-Loading Efficiency of the Films of pHEMA and p(HEMA-co-PEG-MA)-1 to p(HEMA-co-PEG-MA)-3 and the Amounts of Albumin and Fibrinogen Adsorbed onto the Films of pHEMA and p(HEMA-co-PEG-MA)-1 to p(HEMA-co-PEG-MA)-3 After an 18-h Incubation Period at 37°C

Type of film	Loading efficiency (%)	Albumin (ng/cm ²)	Fibrinogen (ng/cm ²)
pHEMA	72	1417 \pm 13	369 \pm 11
p(HEMA-co-PEG-MA)-1	78	223 \pm 13	78 \pm 7
p(HEMA-co-PEG-MA)-2	85	208 \pm 11	63 \pm 6
p(HEMA-co-PEG-MA)-3	94	178 \pm 9	46 \pm 4

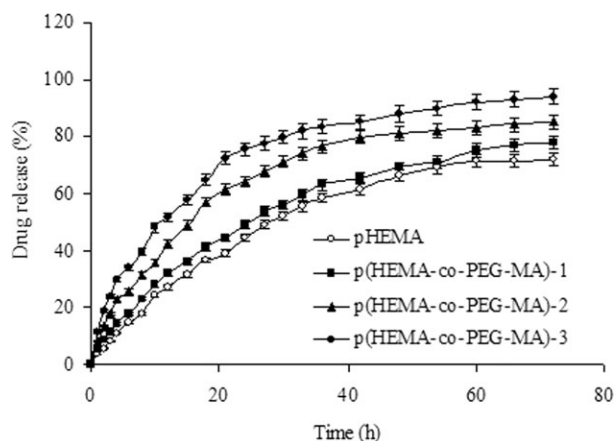


Figure 5 Percentage release of minocycline from the films of the control pHEMA and p(HEMA-co-PEG-MA)-1 to p(HEMA-co-PEG-MA)-3.

chains should have been moved from the interior to the surface of the materials because the material bulk should have been less hydrophilic than that of the aqueous surroundings, so all PEG introduced into the films showed good antifouling surface properties. There were no significant differences in the levels of protein adsorption as a function of the PEG content of the rods, as observed in studies of the water content and contact angles. These results were in good agreement with the related literature.^{18–23}

In vitro drug-release studies

The results for the minocycline loading efficiency of the hydrogels are presented in Table III. The entrapment efficiency (%) of a drug depends on the type of matrix material, the entrapment method of the drug, and the preparation conditions. As can be seen in the table, the drug-loading efficiency of the films depended on the comonomer ratio. The entrapment efficiency increased from 72 to 94% when the PEG-MA comonomer ratio in the polymer network was decreased. As the HEMA/PEG-MA ratio decreased, the minocycline content in the film increased. The effect of the comonomer ratio on the minocycline release kinetics for the p(HEMA-co-PEG-MA) films is presented in Figure 5. The release profiles indicate that the amount of minocycline released decreased with the HEMA/PEG-MA ratio of the hydrogel increasing. The 1 : 2 HEMA/PEG-MA ratio films showed 94% release, whereas the 2 : 1 HEMA/PEG-MA ratio films showed 78% release at 37°C and 72 h. These results should be attributed to the more hydrophilic nature of p(HEMA-co-PEG-MA)-3 formulations (see Table I). With the amount of the comonomer (i.e., PEG-MA) increasing in the copolymer, cumulative release from the formulation was increased. This observation was also expected

because more hydrophilic PEG in the copolymer structures resulted in faster movement of the water to the surface of the drug-loaded film.²²

Kinetic evaluation of drug release

The mathematical theory of diffusion in isotropic substances is based on the hypothesis that the rate of transfer of the diffusing substance through a unit of area of a section is proportional to the concentration gradient measured normal to the section (i.e., Fick's first law of diffusion). Fick's first law assumes that the concentration gradient is independent of time. However, during a real diffusion process, the concentration of the diffusing species within the host solid is changing with the time at any given position. The Fickian mechanism is often applied to diffusion-controlled release behavior because of its simple mathematics. A situation that does not conform to these conditions is described as either case II or anomalous behavior.²⁴ The effect of simple geometry on drug release is described by a power-law model, which is a very suggestive tool for studying the drug-release mechanism dependent on the n value:

$$M_t = M_\infty K_p t^n \quad (3)$$

where M_t and M_∞ are the respective masses of the drug released at time t and infinite (equilibrium) time, respectively; n is the diffusion exponent and is related to the drug-release kinetics; and K_p is the kinetic constant, which incorporates the structural and geometric characteristics of the device and can be explained by the difference in the swelling behavior depending on the polymer composition of the disks. Information about the release mechanism can be gained if we fit the drug-release data and compare the value of n to the semiempirical value of various geometries reported by Peppas and Ritger:²⁵

$$M_t/M_\infty = 4/l(Dt/\pi)^{0.5} \quad (4)$$

where l and D are the film thickness and the apparent diffusion coefficient, respectively. A plot of M_t/M_∞ versus $(t/l)^{0.5}$ is initially linear, and from the slope of $2(D/\pi l^2)^{0.5}$, D can be evaluated.

An attempt was made initially to characterize the experimental release from the hydrogels with the simple Higuchi model.²⁶ The Higuchi model predicts that the release rate will be proportional to the square root of time as follows:

$$Q = k_H t^{1/2} \quad (5)$$

where Q is the mass of drug release per unit of surface area at time t and k_H is Higuchi's release rate constant.

TABLE IV
Minocycline Release Kinetic Models (Power-Law, Zero-Order, and Higuchi Equations) from the Films of pHEMA and p(HEMA-co-PEG-MA)-1 to p(HEMA-co-PEG-MA)-3

Film sample	Release kinetic model							
	Power-law model				Zero-order model		Higuchi model	
	K_p (h^{-n})	n	R^2	$D \times 10^9$ ($\text{cm}^2 \text{s}^{-1}$)	k_0 (mg/h)	R^2	k_H ($\text{mg cm}^2 \text{h}^{-1/2}$)	R^2
pHEMA	0.055	0.733	0.997	0.977	0.206	0.953	0.028	0.992
p(HEMA-co-PEG-MA)-1	0.077	0.639	0.995	0.859	0.208	0.952	0.029	0.993
p(HEMA-co-PEG-MA)-2	0.114	0.559	0.994	0.814	0.215	0.905	0.030	0.974
p(HEMA-co-PEG-MA)-3	0.152	0.484	0.991	0.697	0.218	0.893	0.031	0.976

k_0 , the zero order release rate constant.

The goodness of fit in different release kinetic models (power-law, zero-order and Higuchi models) was evaluated to understand the mechanism of drug release (Table IV). The high values of the coefficient of linear regression (≥ 0.97) for the power-law and Higuchi models confirmed that the data treatment could be used validly for hydrogel films. The correlation coefficients for the power-law model were higher than those for the Higuchi model for all films. As shown in Table IV, the n values related to drug-release kinetics were in all cases lower than 1, and this indicated that drug release was not time-dependent and controlled by the relaxation process due to the swelling of the polymeric network. Drug-release mechanisms of the polymeric films were also evaluated with the Korsmeyer-Peppas semiempirical model.²⁷ In this model, the value of n identifies the release mechanism of the drug; $0.45 \leq n$ corresponds to a Fickian diffusion mechanism, $0.45 < n \leq 0.89$ corresponds to non-Fickian transport, $n = 0.89$ corresponds to case II (relaxational) transport, and $n > 0.89$ corresponds to super case II transport.^{28,29} The values of release parameters n and k (rate constant) are inversely related. A higher value of k may suggest burst drug release from the polymeric matrix. The minocycline release mechanism from hydrogel films of p(HEMA-co-PEG-MA)-1 to p(HEMA-co-PEG-MA)-3 was evaluated, and the n values were observed to be between 0.639 and 0.484 (Fig. 6 and Table V). The *in vitro* release mechanism in the films was therefore taken to be following a Fickian diffusion mechanism. Therefore, the carrier implant in the form of a hydrogel has a predictive ability for minocycline release. Films exhibiting the Higuchi model release mechanism could be obtained with different amounts of the drug-loaded formulation (Table V). However, a zero-order release mechanism could not be found to be valid for different hydrogels. The D values of minocycline from the films were analyzed with eq. (5), and the results are given in Table V. The D values of the films of p(HEMA-co-PEG-MA)-1 to p(HEMA-co-PEG-MA)-3 changed between 0.859×10^{-9} and $0.697 \times 10^{-9} \text{ cm}^2/\text{s}$. D

decreased from 0.859×10^{-9} to $0.697 \times 10^{-9} \text{ cm}^2/\text{s}$ with the PEG-MA content increasing in the films.

Figure 7(a-c) shows the effect of the drug loading on the release pattern as a function of time for films of p(HEMA-co-PEG-MA)-1 to p(HEMA-co-PEG-MA)-3. In all the samples, at any given time, the drug release increased with an increase in the percentage loading of the minocycline in films at pH 7.4 and at 37°C. As shown in Figure 7, the release of minocycline from each hydrogel formulation was nearly linear in the first 30 h, and this suggested an almost constant rate of minocycline release during this period. The hydrogel formulations had similar release profiles, despite different minocycline contents. This indicates that at a lower concentration of the drug, most of the drug molecules remained bound to the polymeric chains of the hydrogels, whereas at a higher concentration of the drug in the hydrogels, the percentage of unbound drug molecules increased in the polymer networks. The maximum minocycline release for the hydrogel films with the 1 : 2 HEMA/PEG-MA ratio [i.e., p(HEMA-co-PEG-MA)-3] with different drug amounts loaded (5, 10, 15, and 20 mg/mL) was determined to be 43, 71, 85, and 94% at 72 h, respectively. Minocycline is freely soluble in a physiological phosphate buffer (pH 7.4); as a result, the rate of depletion of the

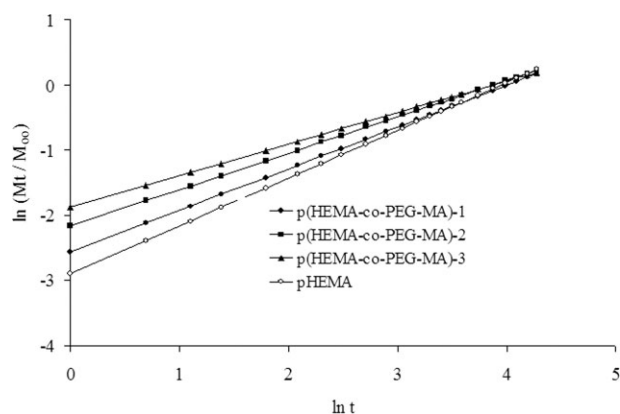


Figure 6 Kinetic evaluation of minocycline release from the hydrogels by the power-law model.

TABLE V
Drug-Release Mechanisms of the Minocycline-Loaded Films of p(HEMA-co-PEG-MA)-1 to p(HEMA-co-PEG-MA)-3 Evaluated with the Korsmeyer-Peppas Semiempirical Model

Film sample	Drug (mg/mL)	Release kinetic model							
		Power-law model				Zero-order model		Higuchi model	
		K_p (h^{-n})	n	R^2	$D \times 10^9$ ($\text{cm}^2 \text{s}^{-1}$)	k_0 (mg/h)	R^2	k_H ($\text{mg cm}^2 \text{h}^{-1/2}$)	R^2
p(HEMA-co-PEG-MA)-1	5	0.049	0.763	0.989	0.995	0.086	0.942	0.012	0.988
	10	0.058	0.720	0.991	0.938	0.147	0.949	0.020	0.992
	15	0.067	0.685	0.989	0.880	0.180	0.939	0.025	0.989
	20	0.077	0.639	0.995	0.859	0.208	0.952	0.029	0.993
p(HEMA-co-PEG-MA)-2	5	0.048	0.754	0.994	0.956	0.105	0.953	0.014	0.989
	10	0.064	0.692	0.998	0.945	0.166	0.956	0.023	0.993
	15	0.073	0.668	0.995	0.962	0.216	0.934	0.029	0.984
	20	0.114	0.559	0.990	0.814	0.215	0.905	0.030	0.974
p(HEMA-co-PEG-MA)-3	5	0.051	0.769	0.988	1.069	0.126	0.919	0.018	0.974
	10	0.081	0.654	0.986	0.958	0.195	0.908	0.027	0.972
	15	0.118	0.549	0.992	0.804	0.214	0.911	0.030	0.975
	20	0.152	0.484	0.989	0.697	0.218	0.893	0.031	0.976

k_0 , the zero order release rate constant.

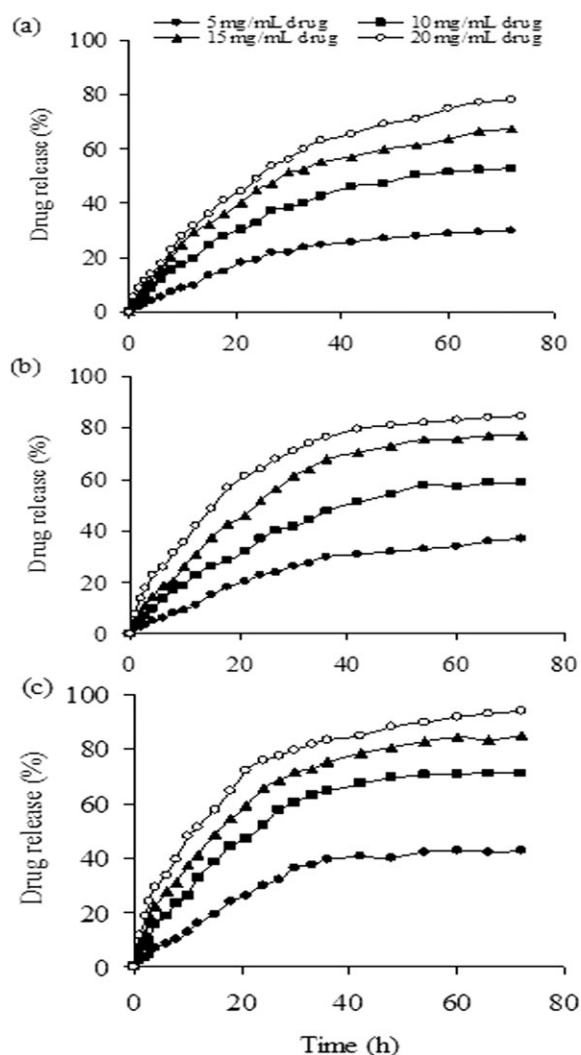


Figure 7 Percentage release of minocycline from the (a) p(HEMA-co-PEG-MA)-1, (b) p(HEMA-co-PEG-MA)-2, and (c) p(HEMA-co-PEG-MA)-3 films loaded with different amounts of the drug.

drug in the polymer at a high concentration was faster than at a low concentration. In this process, the hydrogel became more permeable at a high drug concentration, and this increased the diffusion constant of the drug in the polymer network.^{30,31} In Table V, it can be observed that the diffusion constant of the drug was much lower for the 5 mg/mL drug-loaded p(HEMA-co-PEG-MA)-3 films than for the 20 mg/mL drug-loaded p(HEMA-co-PEG-MA)-3 films.

CONCLUSIONS

In this study, novel wound-dressing and drug-release hydrogel films were fabricated with the HEMA monomer and PEG-MA macroinimer. Both the water contact angles and water content measurements demonstrated that the hydrophilicity of the materials was increased by the introduction of macroinimer PEG-MA into the copolymer structure because the hydrophilic property of a biomaterial is an important parameter for mimicking the moisture of soft tissue. From the DSC curves, pure pHEMA was found to be more thermally stable than p(HEMA-co-PEG-MA)-3 compositions. The protein adsorption studies demonstrated that the introduction of PEG-MA into the pHEMA structure significantly improved the biocompatibility of the material by improving antifouling properties to blood proteins. Meanwhile, the amounts of adsorbed HSA and fibrinogen were significantly reduced in comparison with pHEMA. According to the obtained data, the amount of the released drug is dependent on the hydrogel film composition, and in this way, the amount of the released drug can be tailored according to need by the selection of an adequate composition. All these *in vitro* results indicate that

the p(HEMA-co-PEG-MA) films are good candidates for wound-dressing materials with a controlled drug-release system.

References

1. Bayramoglu, G.; Arica, M. Y. *Macromol Symp* 2003, 203, 213.
2. Larsson, A. Z.; Ekblad, T.; Andersson, O.; Liedberg, B. *Biomacromolecule* 2007, 8, 287.
3. Dong, J.; Chen, L.; Ding, Y. M.; Han, W. J. *Macromol Chem Phys* 2005, 206, 1973.
4. Xiang, Y.-Q.; Zhang, Y.; Chen, D.-J. *Polym Int* 2006, 55, 1407.
5. Dai, Y.; Li, P.; Wang, A. *Prog Chem* 2007, 19, 362.
6. Werner, C.; Maitz, M. F.; Sperling, C. *J Mater Chem* 2007, 17, 3376.
7. Toth, R.; Ferrone, M.; Miertus, S.; Chiellini, E.; Fermeglia, M.; Pricl, S. *Biomacromolecules* 2006, 7, 1714.
8. Mohapatra, R.; Ray, D.; Swain, A. K.; Pal, T. K.; Sahoo, P. K. *J Appl Polym Sci* 2008, 108, 380.
9. Das, A.; Ray, A. R. *J Appl Polym Sci* 2008, 108, 1273.
10. Bayramoglu, G.; Yilmaz, M.; Batislam, E.; Arica, M. Y. *J Appl Polym Sci* 2008, 109, 749.
11. Casimiro, M. H.; Gil, M. H.; Leal, J. P. *Nucl Inst Methods Phys Res Sect B* 2007, 265, 406.
12. Gasteier, P.; Reska, A.; Schulte, P.; Salber, J.; Offenhausser, A.; Moeller, M.; Groll, J. *Macromol Biosci* 2007, 7, 1010.
13. Rokhade, A. P.; Patil, S. A.; Belhekar, A. A.; Halligudi, S. B.; Aminabhavi, T. M. *J Appl Polym Sci* 2007, 105, 2764.
14. Jin, J.; Song, M. *J Appl Polym Sci* 2006, 102, 436.
15. Xu, H.; Ma, L.; Shi, H.; Gao, C.; Han, C. *Polym Adv Technol* 2007, 18, 869.
16. Ahuja, M.; Mahendra, B.; Kanwaljit, C. *Toxicology* 2008, 244, 111.
17. van Oss, C. J. *J Mol Recognit* 2003, 16, 177.
18. Ward, R.; Anderson, J.; McVenes, R.; Stokes, K. *J Biomed Mater Res A* 2007, 80, 34.
19. You, Y.-Z.; Oupicky, D. *Biomacromolecules* 2007, 8, 98.
20. Arica, M. Y.; Tuglu, D.; Basar, M. M.; Kılıc, D.; Bayramoglu, G.; Batislam, E. *J Biomed Mater Res B* 2008, 86, 18.
21. Tomic, S. L.; Suljovrujic, E. H.; Filipovic, J. M. *Polym Bull* 2006, 57, 691.
22. Jung, Y. P.; Kim, J.-H.; Lee, D. S.; Kim, Y. H. *J Appl Polym Sci* 2007, 104, 2484.
23. Lakshmi, S.; Jayakrishnan, A. *Artif Organs* 1998, 22, 222.
24. Wu, X. Y.; Eshun, G.; Zhou, Y. *J Pharm Sci* 1998, 887, 5.
25. Peppas, N.; Ritger, P. L. *J Controlled Release* 1997, 45, 35.
26. Losi, E.; Bettini, R.; Santi, P.; Sonvico, F.; Colombo, G.; Lofthus, K.; Colombo, P.; Peppas, N. A. *J Controlled Release* 2006, 111, 212.
27. Serra, L.; Doménech, J.; Peppas, N. A. *Biomaterials* 2006, 27, 5440.
28. Serra, L.; Doménech, J.; Peppas, N. A. *Eur J Pharm Biopharm* 2006, 63, 11.
29. Siepmann, J.; Peppas, N. A. *Adv Drug Delivery Rev* 2001, 48, 139.
30. Brazed, C. S.; Peppas, N. A. *Eur J Pharm Biopharm* 2000, 49, 47.
31. Siepmann, J.; Gopferich, A. *Adv Drug Delivery Rev* 2001, 48, 229.