A New Degradable Hydroxamate Linkage for pH-Controlled Drug Delivery

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Received August 3, 2006; Revised Manuscript Received October 3, 2006

A new drug delivery system based on a hydrodegradable hydroxamate linkage was evaluated. The carrier support system was poly(*N*-hydroxyacrylamide), which was synthesized via free radical polymerization of acryloyl chloride in 1,4-dioxane, initiated with 2,2′-azobisisobutyronitrile. The poly(acryloyl chloride) was modified in two steps. First, *N*-hydroxysuccinimide was added to give the imide ester of poly(acryloyl). In the second step, the imide ester of poly(acryloyl) was reacted with either hydroxylamine or *N*-methylhydroxylamine to give the corresponding hydroxamic acid. The hydroxamide functionality was then used to link the model drug ketoprofen. All products and intermediates were characterized by elemental analysis and FTIR and ^{1}H NMR spectra. In vitro drug release was performed under specific conditions to elucidate the influence of the pH, polymer microstructure, and temperature on the hydrolysis rate of the amido−ester bond that linked the drug to the macromolecule. The drug release rate from *N*-methylhydroxamic acid polymers was faster than from hydroxamic acid polymers. All polymers showed higher rates of drug release at higher pH values (9.0 > 7.4 > 2.0) and at higher temperatures (37 °C > 20 °C).

Introduction

Controlled drug delivery technology represents one of the most rapidly advancing areas of administering pharmaceuticals.[1] The applications of controlled drug delivery include sustained delivery (over days/weeks/months/years) and are targeted to a specific organ or other host location.[2] Polymer-based drug delivery systems are used to optimize the therapeutic properties of drugs and render them safer and more effective and reliable.[3−10] Targeting of drugs to the specific site of their action provides several advantages over nontargeted drugs. The main advantages are the prevention of drug side effects on healthy cells and enhancement of the drug uptake by targeted cells. Side effects, which invariably impose dose reduction, treatment delay, or even discontinuance of therapy, should be avoided especially for modern drugs with exceptionally high specific activity.

Delivery of drugs via the colon offers numerous therapeutic advantages. Drugs, which are destroyed by the stomach acid and metabolized by pancreatic enzymes, are minimally affected in the colon.[11,12] Sustained colon time release of drugs can be useful in the treatment of certain diseases. The successful delivery of drugs to the colon via the gastrointestinal (GI) tract requires the protection of a drug from being released in the stomach and small intestine. This can be achieved by the use of a special drug delivery system (DDS) that can protect the drug during its transfer to the colon. On the other hand, the drug must be released in the colon from the DDS. Several approaches have been developed for targeted colon drug delivery. Most of them utilize the following four main properties of the GI tract and colon: (1) approximation of the transit time of the small intestine, (2) different physiological conditions in different branches of the GI tract, (3) specificity of bacterial enzymes localized in the colon, (4) targeting of DDS to the colon utilizing targeting moieties specific to the colon.[11]

Hydroxamic acids are known for their ability to form complexes with heavy metals, particularly iron(III).[13−21] A number of hydroxamic acids exhibit biological activity, such as urease inhibition[22−26] and anticoagulant activity.[27] Hydroxamic acid polymers have been investigated as a protective coating for implantable medical devices[28] or as a polymeric drug for the treatment of urinary stones associated with urea-splitting bacteria.[23] In general, hydroxamic acid and its derivatives are relatively nontoxic and biocompatible. Because of their unique characteristics, hydroxamic acid-functionalized polymers have important applications.[13]

Generally, polymers bearing hydroxamic acid groups were synthesized by either polymerization of vinyl monomers bearing hydroxamic acid groups or by converting labile functional groups on a polymer into the corresponding hydroxamide. Domb reported using poly(acrylamide)s as starting materials for the synthesis of hydroxamic acid polymers by reacting the polymers with hydroxylamine at room temperature under basic conditions.[22] Becke and Mutz reported the preparation of acrylohydroxamide from ethyl acrylate,[27] while Narita et al.[30] prepared methacrylohydroxamide from the corresponding ester. Coffman[31] and later Cocea et al.[32] treated maleic anhydride copolymers with hydroxylamine. Treatment of polyacrylonitrile with hydroxylamine followed by hydrolysis of the intermediate amidoxime attached pendent hydroxamic acid groups.[33−36] The acid chloride method that includes treating the poly(acryloyl chloride) with hydroxylamine in dimethylformamide was used by Vrancken and Smets.[37] Generally, the reaction of esters with hydroxylamine proceed normally, and a standard procedure for

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10.1021/bm060755j CCC: $37.00
Published on Web 11/29/2006

preparing hydroxamic acids has been used in a number of instances for preparing polymers.[38,39] Kern and Schultz reported the reaction of poly(methyl acrylate) with hydroxylamine to form a polymer containing 80% acrylohydroxamic acid, 14% acrylic acid, and 6% methyl acrylate.[38]

In the current work, a new method for the synthesis of hydroxamic and *N*-methylhydroxamic acid polymers was developed starting with poly(acryloyl chloride). The resulting polymers bearing hydroxamic and *N*-methylhydroxamic acid groups were used as a polymeric drug carrier system for ketoprofen. The use of hydroxamic and *N*-methylhydroxamic acid polymers as a drug delivery system represents a new approach for special drug delivery that can protect the drug during its transfer to the colon. The dependence of the release of the drug from the polymer on the polymer microstructure, temperature, and pH of the release medium was investigated.

Experimental Section

Materials. Hydroxylamine hydrochloride, 1,3-dicyclohexylcarbodiimide (DCC), tetrahydrofuran (THF), sodium phosphate dibasic (Na_2HPO_4), and potassium phosphate monobasic (KH_2PO_4) were purchased from Aldrich. *N*-Methylhydroxylamine hydrochloride and *N*-hydroxysuccinimide (NHS) were purchased from Acros. Ketoprofen was purchased from Sigma. Acryloyl chloride (AC) was purchased from Aldrich and was distilled at atmospheric pressure before use. 2,2′-Azobisisobutyronitrile (AIBN) was purchased from Aldrich and was recrystallized from methanol. Triethylamine (TEA) (from Acros) was distilled before use. *N*,*N*-Dimethylformamide (DMF) (from Acros) was distilled under vacuum and then stored over molecular sieves. 1,4-Dioxane, purchased from Acros, was distilled and stored over molecular sieves.

Characterization Techniques. FTIR spectra were recorded on an FT-Roman module, MAGNA-IR 760 spectrometer (Nicolet); the samples were examined as KBr disks. ^{1}H NMR spectra were recorded on a Varian Mercury 300 ^{1}H NMR spectrometer. Elemental microanalysis was performed at Quantitative Technologies Inc., Whitehouse, NJ 08888-0470. UV spectra were recorded using a GENESYS 6 spectrophotometer. Gel permeation chromatography (GPC) technology was used to determine the molecular weight of the polymers; a TrisSEC GPC column, eluted with THF, was used with a GPC−viscometery module triple-detector array (TDA), model 300 (Viscotek).

Preparation of Phosphate Buffer (PB) Solution. A pH 9.0 buffer solution was prepared by dissolving Na_2HPO_4 (35.49 g) in 1 L of deionized water; the pH was adjusted to 9.0 using 0.1 N sodium hydroxide and 0.1 N hydrochloric acid solutions. A pH 7.4 buffer solution was prepared by dissolving Na_2HPO_4 (21.70 g) and KH_2PO_4 (2.60 g) in 1 L of deionized water; the pH was adjusted to 7.4 using 0.1 N sodium hydroxide and 0.1 N hydrochloric acid solutions. A pH 2.0 phosphate buffer solution was made from 0.1 N hydrochloric acid, and the pH was adjusted to 2.0 using 0.1 N sodium hydroxide and 0.1 N hydrochloric acid solutions.

Free Radical Polymerization of AC (I). I (23.95 g, 264.62 mmol) in 24 mL of dry 1,4-dioxane in a 100 mL round-bottomed flask was added AIBN (0.25 g, 1.04 wt % AC). The reaction mixture was purged with nitrogen gas for 10 min. The flask was sealed with a rubber septum and was heated at 65−70 °C for 18 h. The poly(acryloyl chloride) (PAC) (**II**) was kept in 1,4-dioxane at room temperature till use.

Poly(*N*-acryloxysuccinimide) (PNAS) (III) Synthesis. To a solution of NHS (15.26 g, 132.59 mmol) in 400 mL of THF in a 500 mL round-bottomed flask was added TEA (13.66 g, 134.99 mmol). The solution was cooled to −5 °C, and then **II** (9.69 g, 107.13 mmol) in 30 mL of a 1:2 mixture of 1,4-dioxane/THF was added dropwise over a 30 min period with vigorous stirring. The reaction mixture was kept at −5 °C for 3 h and at room temperature for 18 h with constant stirring. During this time **III** formed and precipitated from solution. The polymer was filtered and then washed with water and methanol, respectively. After the polymer was dried under vacuum at room temperature for 2 days, 13.05 g (72.06%) of **III** was obtained (Scheme 2). Polymer **III** was characterized using ^{1}H NMR, FTIR, and elemental analysis.

Synthesis of Polymers Bearing Hydroxamide Groups. *Poly(N-hydroxyacrylamide) (PNHA) (**IV**) Synthesis*. To a solution of hydroxylamine hydrochloride (1.97 g, 28.35 mmol) in 15 mL of DMF in a 50 mL round-bottomed flask was added TEA (2.87 g, 28.36 mmol). The mixture was stirred for 30 min, and the precipitated triethylamine hydrochloride formed was removed by filtration. The filtrate containing hydroxylamine was added to a solution of **III** (4.00 g, 23.65 mmol) in 30 mL of DMF in a 100 mL round-bottomed flask. The mixture was stirred at room temperature for 3 days. During this time hydroxamide polymer **IV** formed and precipitated. The hydroxamide polymer **IV** was filtered, washed with water and methanol, respectively, and dried under vacuum at room temperature overnight to yield 1.65 g (80.49%). Polymer **IV** was characterized using ^{1}H NMR, FTIR, and elemental analysis.

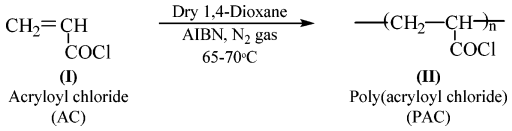
*Poly(N-hydroxy-N-methylacrylamide) (PNHMA) (**V**) Synthesis*. To a solution of *N*-methylhydroxylamine hydrochloride (2.71 g, 32.45 mmol) in 15 mL of DMF in a 50 mL round-bottomed flask was added TEA (3.29 g, 32.51 mmol). The mixture was stirred for 30 min, and the precipitated triethylamine hydrochloride formed was removed by filtration. The filtrate, containing *N*-methylhydroxylamine, was added to a solution of **III** (5.00 g, 29.57 mmol) in 35 mL of DMF in a 100 mL round-bottomed flask. The reaction mixture was stirred at room temperature for 6 days. The solution was concentrated on a rotovaporator at 85−90 °C to one-fourth the volume. Polymer **V** was precipitated by dropwise addition to an excess of acetone (∼60 mL) and recovered by filtration. The product then was dissolved in methanol, filtered, and reprecipitated by the addition of an excess of acetone (400 mL). The product was recovered by vacuum filtration and dried under vacuum at room temperature overnight to yield 1.65 g (55.37%) (Scheme 3). Polymer **V** was characterized using ^{1}H NMR, FTIR, and elemental analysis.

*Poly(N-hydroxyacrylamide)−Ketoprofen Adduct (**VII**) Synthesis*. A mixture of hydroxamide polymer **IV** (0.50 g, 5.75 mmol) and ketoprofen (**VI**) (1.60 g, 6.29 mmol) was mixed with 20 mL of DMF in a 50 mL round-bottomed flask. The mixture was cooled to −5 °C, and a solution of DCC (1.42 g, 6.88 mmol) in 15 mL of DMF was added dropwise over a 20 min period with vigorous stirring. The reaction mixture was stirred at −5 °C for 3 h and at room temperature for 7 days. The precipitated dicyclohexylurea that formed was removed by filtration, and the filtrate was concentrated on a rotovaporator at 80 °C to one-fourth the volume. The polymer−ketoprofen adduct was precipitated by the addition to 50 mL of methanol. Product **VII** was recovered by filtration and dried under vacuum at room temperature for 3 days; the yield was 0.91 g (49.19%) (Scheme 4). The polymer−ketoprofen adduct was characterized using ^{1}H NMR, FTIR, and elemental analysis.

*Poly(N-hydroxy-N-methylacrylamide)−Ketoprofen Adduct (**VIII**) Synthesis*. To the hydroxamide polymer **V** (0.50 g, 4.95 mmol) in 20 mL of DMF in a 100 mL round-bottomed flask was added **VI** (1.38 g, 5.46 mmol). The mixture was cooled to −5 °C, and a solution of DCC (1.12 g, 5.43 mmol) in 10 mL of DMF was added dropwise over a 15 min period with vigorous stirring. The reaction mixture was stirred at −5 °C for 3 h and at room temperature for 5 days. The precipitated dicyclohexylurea that formed was removed by filtration, and the filtrate was concentrated on a rotovaporator at 80 °C until one-fourth the volume. The polymer−ketoprofen adduct was precipitated by dropwise addition to water (200 mL) with rapid stirring. Product **VIII** was recovered by vacuum filtration and washed with acetone. It was dried under vacuum at room temperature for 3 days to give 0.71 g (42.77%) (Scheme 4). The product **VIII** was characterized using ^{1}H NMR, FTIR, and elemental analysis.

Determination of the Molecular Weight of Poly(acryloyl chloride). The molecular weight of **III** was determined by GPC as PAA

Scheme 1. Free Radical Polymerization of AC in 1,4-Dioxane



after complete hydrolysis by treatment of a PNAS solution with 6 N HCl at 100 °C for 24 h.[40] GPC analysis of PAA was performed on a GPC−viscometery module. Polystyrene was used as the standard, and THF was used as the solvent. The M_w of the PAA was 10140 and g/mol, and the M_n was 2246 g/mol with a polydispersity of 4.51.

Determination of the Total Ketoprofen Content. A 10 mg sample of ketoprofen−polymer adduct was suspended in 10 mL of pH 9.0 sodium hydroxide. The mixture was maintained at 60 °C, and the amount of ketoprofen released by hydrolysis was determined with a UV spectrophotometer at λ_{max} = 260 nm on the basis of a standard curve developed with a known amount of ketoprofen.

In Vitro Drug Time Release. The release of ketoprofen was determined with a UV spectrophotometer at λ_{max} = 260 nm as a function of time. The procedure used was as follows: A 100 mg sample of polymer−drug adduct was placed in a 25 mL vial containing 20 mL of phosphate-buffered solutions of pH 2.0, 7.4, and 9.0 and kept at 37 °C (or at 20 °C). At specific intervals, 0.5 mL was collected for analysis, and each time this volume was replaced by fresh medium. Each experiment was carried out in triplicate.

Results and Discussion

Polymer-based drug delivery systems are used to optimize the therapeutic properties of drugs and render them safer and more effective and reliable.[41] Moreover, polymeric drugs and macromolecules when used as drug carriers can be easily synthesized, freely water-soluble, and nontoxic.[42,43] One important goal in drug delivery is that the attached drugs can be targeted to specific areas of the body, tissues, or cells. This method decreases the toxic side effects of the drugs as well as achieves controlled drug delivery.[44] In this work, a new controlled drug release system based on the use of hydroxamic acid polymers is discussed.

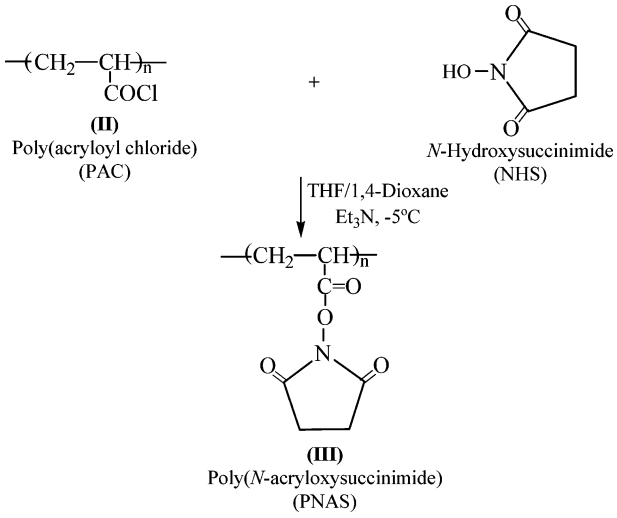
Hydroxamic and *N*-Methylhydroxamic Acid of Poly(acryloyl chloride). Modified poly(acrylamide) polymers are suitable candidates for biomaterial applications due to their facile synthesis and functionalization by the introduction of hydrophobic/hydrophilic moieties.[45] The utilization of poly(acryloyl chloride) in the synthesis of functionalized polymers has been reported.[45] Poly(*N*-substituted acrylamide) and hydroxamic and *N*-methylhydroxamic acid polymers were synthesized in three steps.

Free radical polymerization of distilled acryloyl chloride in 1,4-dioxane initiated with 2,2′-azobisisobutyronitrile gave soluble linear **II** as shown in Scheme 1.

Poly(acryloyl chloride) was then converted to NHS to provide a nucleophilically activated polymer, **III**, as indicated in Scheme 2.[46] For the preparation of hydroxamic acid polymer, **III** was used instead of **II** to enhance the yield reported by Winston et al. (40%) using the latter method.[13] The polymer was soluble in DMF and DMSO but insoluble in water, methanol, 1,4-dioxane, and THF.

The elemental analysis data of **III** are listed in Table 1. On the basis of the nitrogen analysis, polymer **III** contained 84.18% succinimide as depicted (i.e., 84.18% acrylosuccinimide) and 15.82% acrylic acid. The FTIR spectrum of **III** showed a strong peak at 2960 cm^{-1} that was assigned to backbone C−C stretching and strong peaks at 1370 and 1430 cm^{-1} that were assigned to backbone bending. The spectrum had strong peaks at 1740, 1790, and 1820 cm^{-1} that were assigned to amide C═O and ester C═O, respectively. The ^{1}H NMR spectrum of **III** (300 MHz, DMSO-d_6, ppm, from $Si(CH_3)_4$) showed δ 1.40−2.20 (multiplet, CH_2, CH) representing the polymer backbone and δ 2.46 (singlet, CH_2) for the succinimide moiety.

Scheme 2. Chemical Modification of **II** with NHS



Activated **III** was reacted with hydroxylamine.[47] A solution of **III** with hydroxylamine in DMF was stirred at room temperature for 3 days to allow maximum conversion of the imide ester group to form **IV**, which precipitated during the reaction and was soluble in water and DMSO but insoluble in DMF, chloroform, and acetone.

The elemental analysis data for **IV** are listed in Table 1. On the basis of the nitrogen analysis, the conversion was 67.10%. This indicated that the polymer obtained contained 56.49% structure **IV** (*N*-hydroxyacrylamide), 27.69% structure **III** (acrylosuccinimide), and 15.82% acrylic acid. The FTIR spectrum of **IV** showed a strong peak at 2940 cm^{-1} which was assigned to backbone stretching and strong peaks at 1410 and 1460 cm^{-1} due to backbone bending. The broad peaks at 3230 and 3480 cm^{-1} were due to NH and OH, respectively, and the strong peak at 1690 cm^{-1} was assigned to amide C═O. The ^{1}H NMR spectrum of **IV** (300 MHz, DMSO-d_6, ppm, from $Si(CH_3)_4$) showed δ 1.20−2.20 (multiplet, CH_2, CH), δ 3.0 (singlet, OH), and δ 4.10 (singlet, NH).

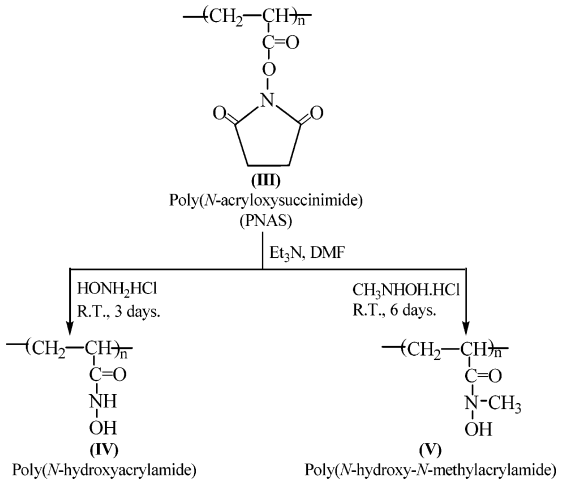
III was reacted with *N*-methylhydroxylamine as shown in Scheme 3. *N*-Methylhydroxylamine hydrochloride was neutralized with triethylamine, and the triethylammonium chloride formed was removed by filtration to give free *N*-methylhydroxylamine that was soluble in DMF. **V** was formed at room temperature by stirring a solution of **III** with *N*-methylhydroxylamine in DMF for 6 days to allow maximum conversion of the imide ester group to an *N*-methylhydroxamic acid group. Product **IV** was precipitated by dropwise addition to acetone.

Table 1. Elemental Microanalysis of Hydroxamic Acid Derivatives of Poly(acryloyl chloride)

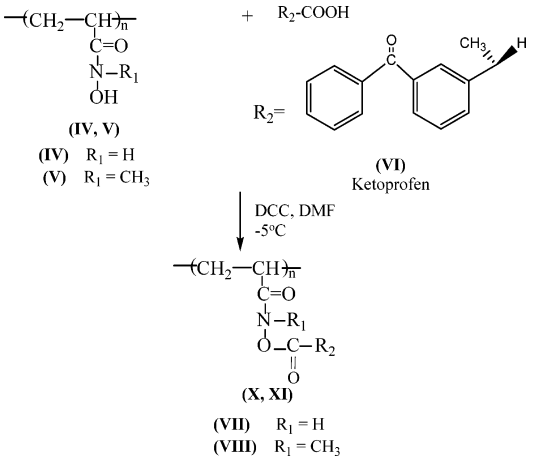
	calculated[a] (%)			found (%)			
polymer	C	H	N	C	H	N	conversion (%)
III	49.7	4.1	8.3	47.9	4.4	7.0	84.0
IV	42.5	5.7	13.9	45.6	5.9	9.3	67.0
V	47.8	6.8	12.2	50.1	7.1	6.5	53.0
VII	65.5	5.1	4.9	64.6	5.7	5.3	99.0
VIII	63.9	5.2	5.2	59.3	7.9	7.8	93.0

[a] The conversion (%) of the starting polymer was considered in the calculations.

Scheme 3. Synthesis of Hydroxamic Acid Polymers



Scheme 4. Immobilization of Ketoprofen onto **IV** and **V**



The polymer was soluble in water, methanol, DMSO, and DMF but insoluble in acetone.

The elemental analysis data for **V** are listed in Table 1. On the basis of the nitrogen analysis, the conversion was 53.27%. This indicates that polymer **IV** contained 44.84% structure **V** (*N*-hydroxy-*N*-methylacrylamide), 39.34% structure **III** (acrylosuccinimide), and 15.82% acrylic acid. The FTIR spectrum of **V** showed a strong peak at 2960 cm^{-1} due to backbone C−C stretching and strong peaks at 1420 and 1460 cm^{-1} due to backbone bending. The strong peak at 1720 cm^{-1} was assigned to amide C═O, and the broad peak at 3460 cm^{-1} was due to OH. The ^{1}H NMR spectrum of **V** (300 MHz, DMSO-d_6, ppm, from $Si(CH_3)_4$) showed δ 1.20−2.20 (multiplet, CH_2, CH), δ 2.48 (singlet, CH_3), and δ 2.92 (broad singlet, OH).

Polymer−Ketoprofen Adduct Synthesis. IV was reacted with ketoprofen in the presence of DCC at −5 °C, to avoid racemization and *N*-acylurea formation,[15] to obtain the polymer−ketoprofen adduct **VII** as indicated in Scheme 4. The reaction was stirred at room temperature for 7 days, and then the precipitated dicyclohexylurea formed was removed by filtration. The polymer−ketoprofen adduct **VII** precipitated by dropwise addition to methanol and was soluble in DMF, DMSO, and acetone and insoluble in water and methanol.

The elemental analysis data for **VII** are listed in Table 1. On the basis of the carbon analysis, the conversion was 99%. This indicated that polymer **VII** contained 56.0% structure **VII** (*N*-hydroxyacrylamide−ketoprofen adduct), 28% structure **IV** (*N*-hydroxyacrylamide), 28% structure **III** (acrylosuccinimide), and 15.8% acrylic acid. Thus, the total amount of ketoprofen loaded onto the polymer−ketoprofen adduct **VII**, based on elemental analysis, was 437.8 (mg of ketoprofen)/(g of polymer). The FTIR spectrum of **VII** showed a strong peak at 2940 cm^{-1} which was due to backbone C−C stretching and a strong peak at 1450 cm^{-1} due to CH_3. The strong peaks at 1660, 1720, and 1800 cm^{-1} were due to amide C═O, ester C═O, and ketone C═O, respectively. The broad peak at 3380 cm^{-1} was assigned to NH. The broad peaks at 3070 and 723 cm^{-1} were assigned to aromatic stretching and bending, respectively. The ^{1}H NMR spectrum of **VII** (300 MHz, DMSO-d_6, ppm, from $Si(CH_3)_4$) showed δ 1.50−2.20 (multiplet, CH_2, CH), δ 1.40 (broad singlet, CH_3), δ 4.30 (broad singlet, NH), and δ 7.3−7.9 (multiplet, H_{arom}).

Table 2. Ketoprofen Content [(mg of ketoprofen)/(g of polymer)] for Polymer−Ketoprofen Adducts **VII** and **VIII** after Sonication for 24 h at 60 °C

polymer	found from hydrolysis	elemental analysis
VII	414.07	438.86
VIII	303.21	313.52

Synthesis of VIII. Activated polymer **V** was reacted with ketoprofen in the presence of DCC at −5 °C to give the polymer−ketoprofen adduct **VIII** as indicated in Scheme 4. The product **VIII** was precipitated in acetone. It was soluble in DMF and DMSO but insoluble in water and acetone. The nitrogen analysis for polymer **VIII** was a little higher than the calculated value as the polymer still had 7% of its structure in the form of the starting polymer, which had higher nitrogen than polymer **VIII**. A similar result was found for polymer **VII**; however, the percentage of polymer **IV** in polymer **VII** is only 1%. Therefore, the amount of nitrogen (%) in polymer **VII** was similar to the calculated value.

The elemental analysis data for **VIII** are listed in Table 1. On the basis of the carbon analysis, the conversion was 93%. The total amount of ketoprofen loaded onto the polymer−ketoprofen adduct **VIII** was found to be 313.5 (mg of ketoprofen)/(g of polymer). The FTIR spectrum of **VIII** showed strong peaks at 2860 and 1450 cm^{-1} which were due to backbone C−C stretching and CH_3, respectively. The strong peaks at 1600, 1630, and 1740 cm^{-1} were due to amide C═O, ester C═O, and ketone C═O, respectively. The broad peak at 3340 cm^{-1} was due to NH, that at 2940 cm^{-1} was due to aromatic stretching, and those at 642 and 727 cm^{-1} were due to aromatic bending. The ^{1}H NMR spectrum of **VIII** (300 MHz, DMSO-d_6, ppm, from $Si(CH_3)_4$) showed δ 0.90−1.30 (multiplet, CH_3, CH_2), δ 1.40−1.90 (multiplet, CH), and δ 7.40−7.90 (multiplet, H_{arom}).

Determination of the Total Ketoprofen Content. To evaluate the total content of ketoprofen in the samples, they were hydrolyzed by heating or sonication using a known amount of the polymer−ketoprofen adduct in alkaline solution of pH 9.0. The amount of ketoprofen released from the sample was determined with a UV spectrophotometer at λ_{max} = 260 nm at room temperature within 30 min. Then the samples were heated at 60 °C. The drug concentration was monitored with a UV spectrophotometer until it reached a constant value.

The fast hydrolysis studies showed that the total ketoprofen content of **VII** was 414.07 (mg of ketoprofen)/(g of polymer) (Table 2). At the same time, the total ketoprofen content of **VIII** was 303.21 (mg of ketoprofen)/(g of polymer) (Table 2). The results obtained by the alkaline hydrolysis of the ketoprofen−polymer adducts were compared with those obtained from elemental analysis. The results of these investigation found that both results are comparable.

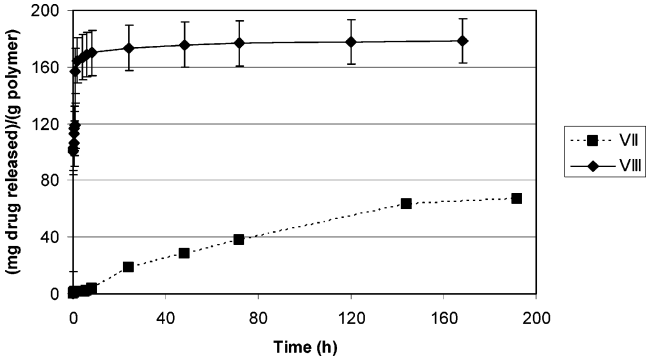


Figure 1. In vitro release profile of ketoprofen from **VII** and **VIII** in phosphate buffer (pH 9.0) at body temperature (37 °C).

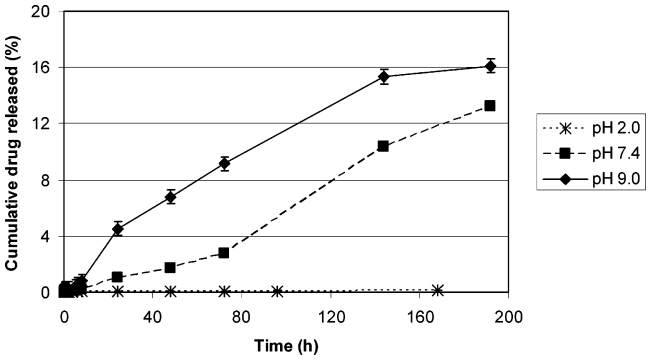


Figure 2. In vitro release profile of ketoprofen from **VII** in phosphate buffer with various pH values at body temperature (37 °C).

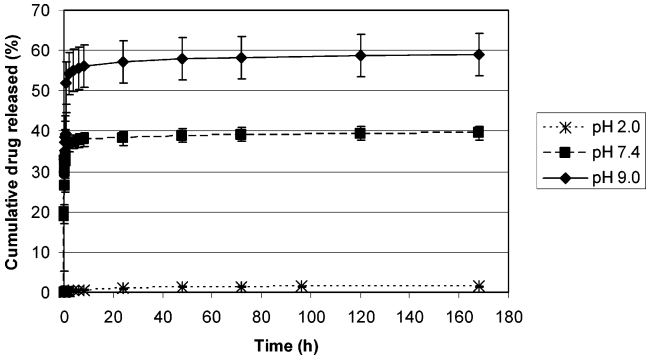


Figure 3. In vitro release profile of ketoprofen from polymer **VIII** in phosphate buffer with various pH values at body temperature (37 °C).

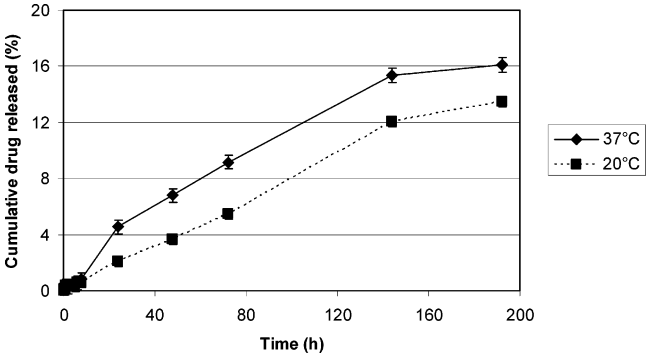


Figure 4. In vitro release profile of ketoprofen from polymer **VII** in phosphate buffer (pH 9.0) at body temperature (37 °C) and at room temperature (20 °C).

In Vitro Drug Release. The rate of ketoprofen released from the polymer formulations was studied at various pH values and temperatures. The rate of release usually depends on the polymer microstructure, the pH of the release medium, and the temperature.

The effect of the polymer microstructure on the release rate of the ketoprofen and polymer−ketoprofen adducts **VII** and **VIII** was studied on the basis of modifications of poly(acryloyl chloride) as shown in Scheme 4. The release rate of the ketoprofen from these polymers containing ketoprofen under various release conditions, such as the pH and temperature of the medium, was investigated. It was found that the polymer−ketoprofen adduct with an *N*-methyl group (**VIII**) showed a faster ketoprofen release rate than the polymer containing NH (**VII**) as shown in Figure 1. At pH 9.0, polymer **VII** showed 16.11% ketoprofen release after 7 days at 37 °C. However, polymer **VIII** released 58.92% of the ketoprofen under the same conditions as shown in Figure 1. Similar results were obtained at 20 °C.

The effect of pH on the release rate of ketoprofen was investigated in specific pH media: pH 2.0 (stomach pH), pH 7.4 (lower GI tract pH), and pH 9.0 (colon pH). It was found that the release rate of ketoprofen increased as the pH increased. In addition, the polymer−ketoprofen adducts **VII** and **VIII** showed high hydrolytic stability in an acidic medium (i.e., within the stomach pH), while the release rate of ketoprofen was greatest in an alkaline medium (i.e., within the colon pH). It was found that polymer **VII** released 0.14% of its drug content after 7 days in a buffer solution of pH 2.0 at 37 °C. However, the release rate of ketoprofen was 14% of its drug content in a buffer solution of pH 7.4, and it released 16% of its drug content in a buffer solution of pH 9.0 under the same conditions as shown in Figure 2; the same trend was observed at 20 °C. The polymer-containing ketoprofen **VII** released about 13.5% of its drug content at pH 9.0 and 4.3% at pH 7.4, and negligible amounts were released at pH 2.0. The total amount of ketoprofen released from polymer **VIII** was 1.5% of its drug content after 7 days in a buffer solution of pH 2.0 at 37 °C. At pH 7.4 the total amount of ketoprofen released was found to be 40% of the drug content, and in a buffer solution of pH 9.0 59% of its drug content was released under the same conditions as shown in Figure 3; the same phenomenon was observed at 20 °C. The release rate was higher at pH 9.0 than at pH 7.4 and 2.0.

To study the effect of the temperature, the release rates of ketoprofen from polymers **VII** and **VIII** were investigated at two temperatures, 37 °C (body temperature) and 20 °C (room temperature). Generally, the aim of this was to investigate the effect of a change in the temperature of the release medium on the release rate, and room temperature was selected because it is lower than body temperature. It was also possible to select a temperature which is higher than body temperature to study the same phenomena. It was found that, by increasing the temperature of the release medium, the release rate of ketoprofen from the polymers was increased.

The total amount of ketoprofen released from polymer **VII** at 37 °C was found to be 0.14% of its drug content after 7 days in a buffer solution of pH 2.0. However, at 20 °C, the total amount of ketoprofen released was found to be 0.03% of its drug content under the same conditions. Similar effects were observed at pH 7.4 and 9.0. At 37 °C, the total amount of ketoprofen released from polymer **VII** was found to be 13% of its drug content after 7 days in a buffer solution of pH 7.4. However, at 20 °C it was found to be 4.3% of its drug content under the same conditions. Moreover, at 37 °C in a buffer solution of pH 9.0, polymer **VII** released 16% of its drug content after 7 days. However, at 20 °C, the total amount of ketoprofen released was found to be 13.5% of its drug content under the same conditions as shown in Figure 4. The total amount of ketoprofen released from polymer **VIII** at 37 °C was found to

be 1.5% of its drug content after 7 days in a buffer solution of pH 2. At 20 °C, polymer **VIII** released 1% of its drug content under the same conditions. The same trend was observed in buffer solutions of pH 7.4 and 9.0. At 37 °C, polymer **VIII** showed a release of 39.6% of its drug content after 7 days in a buffer solution of pH 7.4. At 20 °C, the release rate was found to be 27% of its drug content under the same conditions. At 37 °C, in a buffer solution of pH 9.0, the total amount of ketoprofen released was found to be 59% of its drug content after 7 days. However, at 20 °C, polymer **VIII** released 31% of its drug content under the same conditions.

Conclusions

A new system for oral drug delivery was developed. This system was based on hydroxamic and *N*-methylhydroxamic acid polymers. The hydroxamic and *N*-methylhydroxamic acid groups were used as polymeric drug carriers for ketoprofen. This represents a new approach for the use of hydroxamic and *N*-methylhydroxamic acid polymers since it has a degradable linkage that is dependent on the pH of the environment.

The in vitro release of ketoprofen was determined with a UV spectrophotometer at $\lambda_{max} = 260$ nm as a function of time. The release was monitored in phosphate-buffered solutions of pH 2.0, 7.4, and 9.0 at body temperature (37 °C) and at room temperature (20 °C). The two temperatures were selected to investigate the effect of a change in body temperature. The in vitro release profiles for ketoprofen showed that the amount of drug released from *N*-methylhydroxamic acid polymers was 2.5 times higher than that from hydroxamic acid polymers. The release rate showed a high dependence on the pH and the temperature of the release medium. The polymer−ketoprofen adduct **VII** showed no burst release at all tested pH values. However, the polymer−ketoprofen adduct **VIII** showed burst release at pH 7.4 and 9.0 and no burst release at pH 2.0. The pH dependence of the release of the ketoprofen drug from the polymer−ketoprofen adduct could be in favor of applying these polymeric drugs for targeting the drugs to the colon. Since the polymeric formulation is not highly affected by the low pH of the stomach, it can easily pass to the colon and start releasing the drug at the higher pH of the colon.

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BM060755J