Polyacrylamide-*g*-Alginate-Based Electrically Responsive Hydrogel for Drug Delivery Application: Synthesis, Characterization, and Formulation Development

Raghavendra V. Kulkarni,[1] Chitrali Mallikarjun Setty,[2] Biswanath Sa[3]

[1]*Department of Pharmaceutical Technology, BLDEA's College of Pharmacy, Bijapur 586103, India*
[2]*Department of Pharmaceutics, Saraswati College of Pharmaceutical Sciences, Hyderabad, India*
[3]*Department of Pharmaceutical Technology, Center for Advanced Research in Pharmaceutical Sciences, Jadavpur University, Kolkata 700 032, India*

Received 2 May 2009; accepted 31 July 2009
DOI 10.1002/app.31203
Published online 15 September 2009 in Wiley InterScience (www.interscience.wiley.com).

ABSTRACT: An electrically responsive hydrolyzed polyacrylamide-*grafted*-sodium alginate (H-PAAm-*g*-SA)-based membrane-controlled transdermal drug delivery systems were developed and evaluated. The grafting reaction was confirmed by Fourier transform infrared spectroscopy, elemental analysis, and thermogravimetric analysis. On application of electric stimulus, the swollen H-PAAm-*g*-SA hydrogel was deswelled in the vicinity of electrodes. The drug release was greater in the presence of electric stimulus when compared with passive diffusion, and it was found to be dependent on the applied electric current strength, concentration of H-PAAm-*g*-SA copolymer in the reservoir, and cross-link density of rate-controlling membrane. A pulsatile pattern of drug release was observed when the electric stimulus was switched "on" and "off." The skin histopathology study suggested that, after application of an electrical stimulus, changes were in the structure of stratum corneum. © 2009 Wiley Periodicals, Inc. J Appl Polym Sci 115: 1180–1188, 2010

Key words: graft copolymer; controlled release; electrically responsive; hydrogel; biomaterials; ketoprofen

INTRODUCTION

The hydrogels that can show dramatic changes in their swelling behavior, network structure, permeability, or mechanical strength in response to different internal and external stimuli are being used to develop responsive drug delivery systems.[1] These are termed as "intelligent" or "smart" hydrogels.[2] Such drug carriers can release the required quantity of drug at a right time and right place in the body on application of stimulus and are most useful in mimicking the in vivo pulsatile release of many endogenous hormones.[3,4] Hydrogels are polymeric materials that do not dissolve in water but swell considerably in an aqueous medium. Hydrogel transitions or volume changes occur in response to changing environmental conditions such as temperature, pH, solvent composition, and electric stimuli. Such hydrogels have been studied for applications in a variety of fields, such as in chemical engineering, medicine and pharmaceuticals, food processing, agriculture, and separation techniques.[5–7]

Correspondence to: B. Sa (pharma_75raghu@yahoo.com) and (biswanathsa2003@yahoo.com).

Journal of Applied Polymer Science, Vol. 115, 1180–1188 (2010)

Electrically responsive hydrogels can be prepared from polyelectrolytes and are thus pH and electrically responsive.[8] The electrically responsive hydrogels have been studied for controlled drug delivery applications by several authors.[9–12] Electrically controlled drug delivery may particularly offer unique advantages for providing on-demand release of drug molecules from implants or transdermal drug delivery systems. In addition, electrical control is advantageous for coupling with sensors and microelectronics in feedback-controlled systems.[13] The electrically modulated drug release occurs via a number of different mechanisms, for example, a charged drug migrates toward the oppositely charged electrode or, more commonly, drug release is largely governed by the effects of electric current on hydrogel matrix. Polyanionic hydrogels contact near anode and water exudes at cathode while the reverse phenomenon holds for polycationic hydrogels. Hydrogel contraction and electroosmosis result in ejection of drugs out of the gels on electrical stimulation.[14]

The grafting of polyacrylamide (PAAm) on the sodium alginate (SA) is of great importance to develop new materials combining the properties of both natural and synthetic polymers.[15] Another advantage of this graft copolymer is that this could be converted into ionic form through the hydrolysis of

amide groups resulting in electrical-sensitive copolymer.[16–18] Recently, we have reported the synthesis and characterization of alkaline hydrolyzed polyacrylamide-*grafted*-sodium alginate (H-PAAm-g-SA) copolymer for pH-sensitive oral drug delivery application.[19] As carboxyl functional groups (—COOH) are present on the backbone of PAAm-*g*-SA copolymer, it can be responsive to electric stimulus also. With this in mind, we have further explored the possibilities of developing H-PAAm-*g*-SA-based electrically responsive transdermal systems. This graft copolymer possesses large number of ionizable —COO groups that are responsible for electrical sensitivity and large polymeric side chains that can entrap higher drug loadings that are important in case of transdermal systems and implants. Although various synthetic and natural polymers have been used to develop electrically responsive delivery systems,[9–14] to our knowledge, there is no report on electrically responsive H-PAAm-*g*-SA hydrogel for transdermal drug delivery application.

Ketoprofen is used in musculoskeletal and joint disorders such as ankylosing spondylitis, osteoarthritis, and rheumatoid arthritis. It is readily absorbed from the gastrointestinal tract, and peak plasma concentrations are attained 0.5 to 2 h after a dose, but it causes a certain irritation in the gastrointestinal mucosa and possesses a bitter taste.[20] The shorter plasma half-life (2–3 h) and associated gastric side effects make ketoprofen a good candidate for transdermal delivery.

The aim of this work was to synthesize and evaluate the feasibility of using electrically responsive H-PAAm-*g*-SA hydrogel for transdermal delivery of ketoprofen, a model nonsteroidal anti-inflammatory drug. The PAAm was grafted on the backbone of SA and subsequently, the —$CONH_2$ groups of PAAm were converted to —COO groups to obtain a polyanionic electrically responsive copolymer. The membrane-controlled transdermal drug delivery systems were developed using H-PAAm-*g*-SA hydrogel as drug reservoir, cross-linked poly(vinyl alcohol) (PVA) film as a rate-controlling membrane (RCM) and a polystyrene film laminate as backing layer. The in vitro release of ketoprofen through the excised rat abdominal skin was studied with or without electric stimulus.

EXPERIMENTAL

Materials

Ketoprofen was obtained as a gift sample from Rhone-Poulenc Ltd. (Mumbai, India). SA, PVA (M_w 1,25,000; 98% hydrolyzed), acrylamide (AAm), ammonium persulfate (APS), glutaraldehyde (GA, 25 mass % aqueous solution), poly(ethylene glycol) 200, sodium hydroxide (NaOH), chloroform, and methanol were purchased from S.D. Fine Chemicals (Mumbai, India) and used as received. A polystyrene film laminate (Scotchpak backing 1006) was obtained as a gift sample from 3M Pharmaceuticals Ltd. Double-distilled water was used throughout the study.

The male albino rats (150–250 g) were acclimatized to laboratory conditions for 3 days before the experiments, with adequate food and water *ad libitum*. Animal experimental protocols were approved by our institutional ethics committee, which follows the guidelines of the committee for the purpose of control and supervision of experiments on animals.

The electric stimulus generating device was designed and fabricated to meet the following requirements: voltage range = 0–30 V; current range = 0–50 mA; current type = dc or pulsed dc; on-off ratios = 1:1; two carbon electrodes (0.5 cm^2 area). The device had manual selector switches for dc, the desired on-off ratio, a knob for voltage, and current adjustments. Current flow was read on an ammeter. The device could be operated on an alternating current of 220 V stepped down to 12 V.

The electric stimulus of 2 to 8 mA and duration of 4 h was used during the experiments. The drug release study was carried out with switching on at intervals of 30 min or without switching. The drug release experiments were conducted under various conditions: (a) without electrical stimulus; (b) with a constant electrical stimulus of 2, 4, and 8 mA currents; and (c) with a switching on-off electrical stimulus with 4 mA current.

Synthesis of electrically responsive H-PAAm-*g*-SA copolymer

The H-PAAm-*g*-SA copolymer was synthesized by free radical polymerization as reported earlier.[19] Briefly, 2 g of SA was dissolved in 100 ml of double-distilled water in a 250-ml three-necked round bottom flask and allowed to hydrate for 4 h with continuous purging of slow stream of nitrogen gas. The flask was heated at 75°C, and 0.12 mol of AAm and 0.002 mol of ammonium persulphate were added to SA solution. Polymerization was carried out for 60 min with continuous purging of nitrogen gas. After 60 min, the resulting copolymer was allowed to cool to ambient temperature and the product was poured into excess methanol and kept for 24 h to dewater. The copolymer was then filtered, washed repeatedly with methanol, and dried at 50°C. Mass of the graft copolymer was taken and the percent grafting efficiency was calculated using the following relation[21]:

$$\% \text{ Grafting Efficiency} = \left(\frac{W_1 - W_0}{W_2}\right) \times 100$$

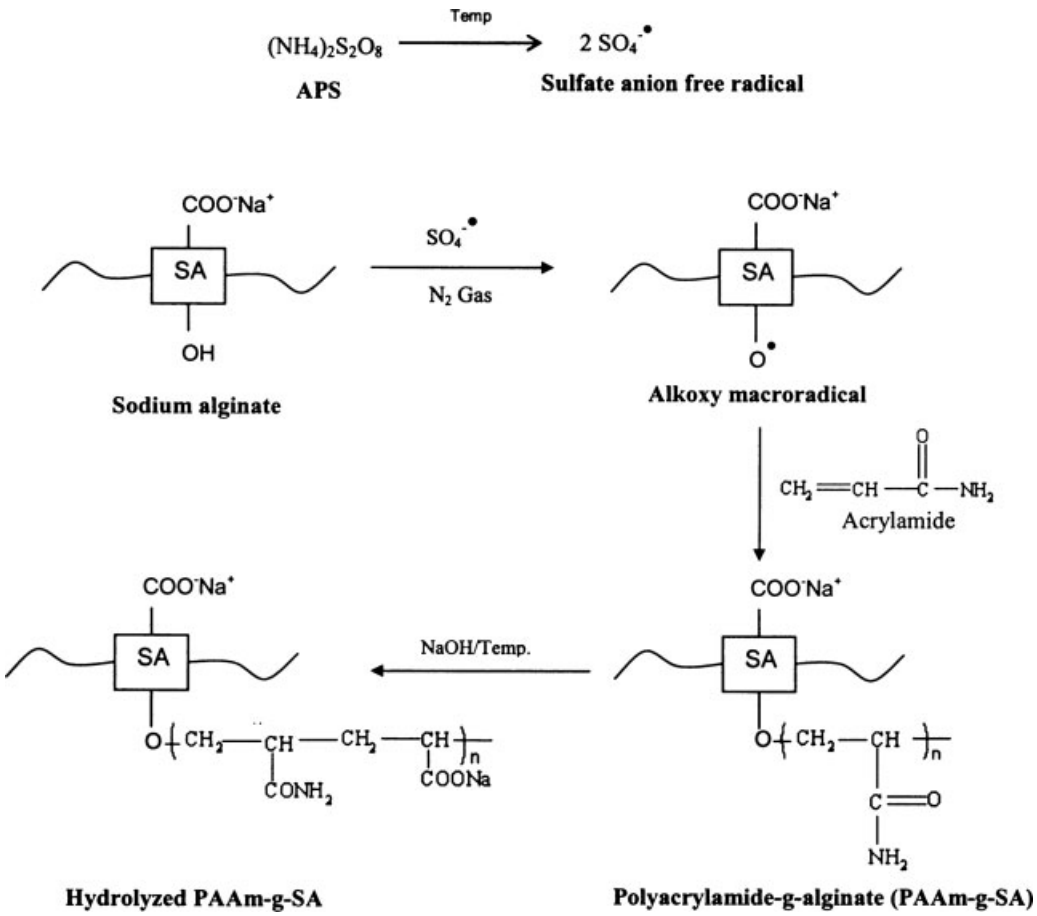

Figure 1 Proposed mechanistic pathway for the synthesis of H-PAAm-*g*-SA copolymer.

where W_0, W_1, and W_2 are the weights of SA, graft copolymer, and AAM, respectively.

Then, the 2 g of PAAm-*g*-SA copolymer was dissolved in 100 ml NaOH solution (0.9*M*) and stirred at 75°C for 60 min in a thermostatic water bath. At the end of the reaction time, the solution was cooled and poured in excess methanol. The hydrolyzed copolymer (H-PAAm-*g*-SA) was separated by filtration and washed repeatedly with methanol and dried overnight at 50°C and stored in a desiccator. The proposed mechanistic pathway for the synthesis of H-PAAm-*g*-SA copolymer is given in Figure 1.

Characterization of copolymer

The synthesized H-PAAm-*g*-SA copolymer was characterized by Fourier transform infrared spectroscopy (FTIR), elemental analysis, and thermogravimetric analysis (TGA).

The FTIR spectroscopy was used to confirm the grafting and alkaline hydrolysis. The samples were crushed with potassium bromide to make pellets under hydraulic pressure of 600 kg. Spectra were taken on a Nicolet (Model Magna 550, USA) instrument and scanned between 500 and 4000 cm^{-1}.

The elemental analysis of the neat SA, PAAm-*g*-SA, and H-PAAm-*g*-SA was done using Flash EA 1112 CHN analyzer (Thermo Finnigan, Italy), and the percent of nitrogen, carbon, and hydrogen were calculated.

TGA was performed on SA and H-PAAm-*g*-SA under a dynamic nitrogen atmosphere flowing at a rate of 50 mL/min and at a heating rate of 10°C/min in the temperature range 0–600°C using a microcalorimeter (DuPont-9900, USA).

Development of drug delivery system

The electrically responsive transdermal drug delivery systems (ETDDS) were prepared by entrapping the hydrogel reservoir within a shallow compartment molded from a backing layer and an RCM.[22] The schematic diagram of the ETDDS is shown in Figure 2 and formulation details are given in Table I.

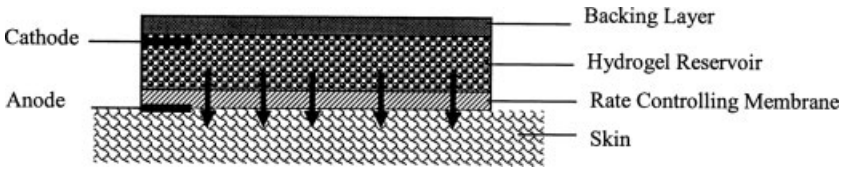

Figure 2 Schematic diagram of the electroresponsive drug delivery system.

The hydrogel reservoir of H-PAAm-*g*-SA was prepared by dissolving known amount of copolymer in double-distilled water using magnetic stirrer for 2 h at 50°C. The 25% (w/w of dry polymer) ketoprofen was uniformly incorporated into polymeric solution; 6% (w/w of dry polymer) GA and 1% of 1N HCl were added, and stirring was continued for 4 h. Finally, the reaction mixture was washed with distilled water and the obtained hydrogel was stored in a well-closed container.

The RCM were prepared by dissolving the known quantity of PVA in distilled water. Poly(ethylene glycol) 200 was added as a plasticizer. This polymeric solution was poured on the mercury surface (28.3 cm^2) in a petri dish and dried at room temperature for 24 h. The obtained membranes were removed from the mercury surface and crosslinked by dipping them into methanol containing GA and 1% of 1*N* HCl for 4 h. The crosslinked membranes were washed with distilled water to remove unreacted GA and dried at room temperature for 24 h.

To prepare ETDDS, an accurately weighed quantity of H-PAAm-*g*-SA hydrogel equivalent to 10 mg of drug was placed on a sheet of backing layer (polystyrene film laminate; 3*M* Scotchpak backing 1006) having an area 1.6 cm^2. An RCM was placed over the hydrogel reservoir and the edges were heat sealed to obtain leak proof device. Then, the prepared ETDDS were stored for 30 days in a well-closed desiccator for further evaluation.

Electrical response of the hydrogel

The electroresponsive property of H-PAAm-*g*-SA hydrogel or neat SA was evaluated under the applied electric stimulus. The 0.5 g of hydrogel was placed inbetween two carbon electrodes (0.5 cm^2) placed 3 cm apart. Then, the electric stimulus of 2, 4, and 8 mA was applied for 30 min. The hydrogel was periodically weighed using electronic microbalance (Model BL-220H, Shimadzu, Japan) having an accuracy of 0.001 mg. The deswelling ratio was calculated using the following relation:

$$\text{Deswelling ratio} = \frac{W_t}{W_o}$$

where W_o is the initial weight of the hydrogel and W_t is the weight of hydrogel at time "t" under electric stimulus.

In vitro drug release experiments

The rats were killed by cervical dislocation just before use. The abdominal skin section was carefully cut, lifted, and separated from the adhering fatty tissues and visceral materials. The hair and underlying subcutaneous dermal fats were scrapped off carefully without damage to the skin. The excised skin was thoroughly washed with double-distilled water, and a skin of 1.7 cm^2 area was used for mounting on the donor compartment of the modified Keshary-Chien diffusion cell, with stratum corneum (SC) side facing the donor compartment.[23]

Vertically assembled modified Keshary-Chien diffusion cells having a downstream volume of 50 ml were used for the study. The receptor compartment was filled with phosphate buffer (pH 7.4), the magnetic stirrer was set at 50 × *g* and whole assembly was maintained at 32 ± 0.5°C. The drug delivery

TABLE I
Composition of Electrically Responsive Drug Delivery Systems

Formulation Codes	Hydrogel reservoir				Rate-controlling membrane		
	H-PAAm-*g*-SA (% w/v)	Neat SA (% w/v)	Drug (%w/w of polymer)	GA (% w/w of copolymer)	PVA (% w/v)	PEG 200 (% w/w of polymer)	GA (% w/w of polymer)
ETDDS1	2	–	25	6	4	10	3
ETDDS2	3	–	25	6	4	10	3
ETDDS3	3	–	25	6	4	10	6
ETDDS4	3	–	25	6	4	10	9
ETDDS5	3	–	25	6	8	10	9
ETDDS6	–	2	25	6	4	10	3

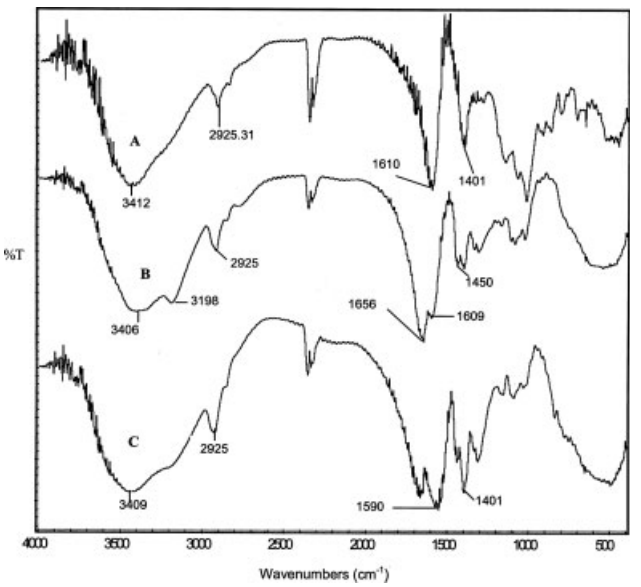

Figure 3 FTIR spectra of sodium alginate (A), PAAm-*g*-SA (B), and H-PAAm-*g*-SA (C).

system was applied on the excised rat skin, which in turn was fixed to the donor compartment. Then, the cathode was placed in the donor compartment and anode was placed in the receptor compartment.[9] The electrical stimulus was then applied to the hydrogel system with regulated DC power source. The amount of drug released was determined by withdrawing 5 ml samples at specific time intervals, volume withdrawn was replaced with equal volume of fresh buffer, and samples were analyzed using UV-visible spectrophotometer (Model Pharmaspec UV-1700, Shimadzu, Japan) at 260 nm.

Histopathological evaluation

The skin samples before and after electrically stimulated drug permeation experiments were washed with normal saline and preserved in 10% buffered formalin solution for histopathological examination.[24] The histopathological study was carried out within 24 h after storage in formalin solution. The tissues were processed according to routine light microscope technique. First, they were dehydrated in ascending degrees of ethyl alcohol (70, 80, 90, 96, and 99%), cleared in xylene, and embedded in paraffin. Five-micrometer-thick paraffin sections were cut and stained with hematoxylin-eosin, examined and photographed under binocular light microscope (CXRIII, Labomed, Mumbai, India).

The statistical analyses were accomplished using Graphpad Instat statistical package. The one-way analysis of variance was used to determine the statistically significant differences between the results. The results with $P < 0.05$ were considered statistically significant.

RESULTS AND DISCUSSION

The grafting of AAm on the backbone of SA was carried out by free-radical polymerization using APS as reaction initiator under nitrogen atmosphere. The reaction temperature was maintained at 75°C; at this temperature, the APS undergoes decomposition to produce sulfate anion free radical, which abstracts the hydrogen from hydroxyl groups of SA to form alkoxy radical on the substrate. Then, the resulting macroradical initiates the graft copolymerization of AAm onto the backbone of SA. The grafting efficiency was found to be 88.37%.

Characterization of the copolymer

Figure 3 shows the FTIR spectra of neat SA, PAAm-*g*-SA, and H-PAAm-*g*-SA. In the case of neat SA, the broad peak appearing at 3412 cm^{-1} corresponds to the presence of hydrogen-bonded OH groups. The peaks appearing at 1610 and ~1400 cm^{-1} are due to the COO^- groups and the peak appearing at 2925 cm^{-1} is due to the C—H stretching. In the spectra of PAAm-*g*-SA, the peaks at 3406 and 3198 cm^{-1} are attributed to the overlap of N—H stretching band of amide group and O—H stretching band of hydroxyl groups of SA. The peaks at 1656 and 1450 cm^{-1} are due to the presence of primary amide group on the backbone of SA. This confirms the grafting reaction. Whereas, in the case of H-PAAm-*g*-SA copolymer, the sharp peaks appearing at 3406 and 3198 cm^{-1} were absent, indicating the absence of N—H band and the peaks at 1590 and 1401 cm^{-1} are due to COO^- groups. This confirms the hydrolysis reaction.

The elemental analyses data are depicted in Table II. A 0% nitrogen, 27.367% carbon, and 4.226% hydrogen were found in neat SA, but a considerable increase in nitrogen (14.275 %) was seen in PAAm-*g*-SA. This could be accounted for the presence of —$CONH_2$ groups on the backbone of SA. On the other hand, the percent nitrogen was reduced to 3.28% in H-PAAm-*g*-SA. After hydrolysis, the —$CONH_2$ groups were partially converted to —COOH groups (partial hydrolysis of 77.03%), leading to reduced nitrogen content, thus confirming the grafting and alkaline hydrolysis reactions.

TGA thermograms of neat SA and H-PAAm-*g*-SA copolymers are shown in Figure 4. The neat SA starts decomposing after 205°C and the mass loss

TABLE II
Elemental Analysis Results

Polymer	% N	% C	% H
Neat SA	0.000	27.367	4.226
PAAm-*g*-SA	14.275	40.899	7.001
H-PAAm-*g*-SA	3.282	38.268	5.190

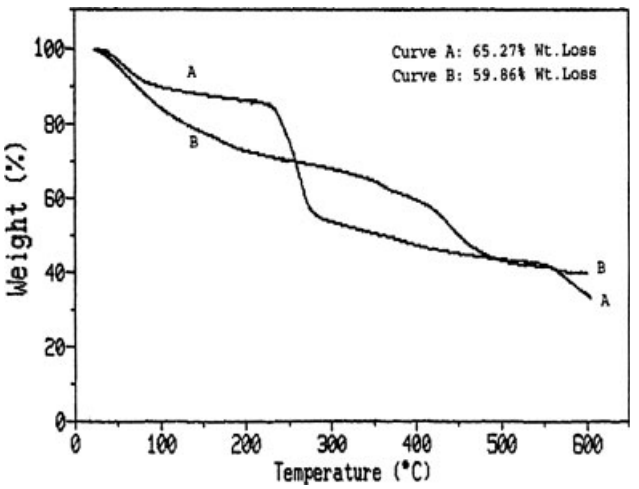

Figure 4 TGA thermograms of sodium alginate (A) and H-PAAm-*g*-SA (B).

(14%) up to 205°C may be due to loss of loose and bound water in the polymer. A sharp mass loss (43%) was observed between 205 and 533°C and reached a value of 65.3% at 600°C. This may be attributed to the decomposition of polymer. In the case of H-PAAm-*g*-SA copolymer, the decomposition started at higher temperature (255°C) and reached value of 59% at 600°C. The mass loss was found to be constant and percent residual mass of H-PAAm-*g*-SA copolymer was higher than neat SA. This suggests that the graft copolymer has been formed and is thermally more stable than the neat SA.

Electrical response of hydrogel

The electrical response of H-PAAm-*g*-SA hydrogel or neat SA as a function of time depending on the applied electric current strength and concentration of H-PAAm-*g*-SA is shown in Figure 5. The deswelling of swollen H-PAAm-*g*-SA hydrogel reservoir was observed when an electric stimulus was applied; as the concentration of H-PAAm-*g*-SA was increased in hydrogel reservoir, the deswelling was increased, which may be due to the presence of more number of ionizable —COO groups. Also, with increased electric current strength, the deswelling increases. On the other hand, the neat SA hydrogel did not show electrical responsiveness compared with H-PAAm-*g*-SA hydrogel as there was no significant deswelling. The remarkable deswelling of the copolymer hydrogel reservoir was observed around the anode that may be attributed to the interaction of polyanion with positive charge of anode. At this time, water inside the hydrogel was squeezed together with the drug because of this shrinkage. This deswelling (squeezing effect) could enhance the release of loaded drugs. The similar result has been reported for anionic polyelectrolyte gel.[25]

When an electrical stimulus is applied on the negatively charged copolymer hydrogel, the counter ion of polyion moves toward negative electrode (cathode), whereas the polyion remains immobile. Also, the free ions in the surrounding solution move toward their counter electrodes and come into hydrogel matrix. Thus, the osmotic pressure of the hydrogel matrix near the positive electrode (anode) increases and becomes larger than that of negative electrode side. Consequently, the osmotic pressure difference occurs within the hydrogel and it is a driving force for releasing of drugs entrapped in the matrix. Another factor that influences the release of loaded drugs from polyelectrolyte hydrogels may be the local pH gradient attributed to water electrolysis.[13]

In vitro drug release

The release of ketoprofen from various formulations with or without the electrical stimulus is shown in Figure 6(A,B). In the absence of electric stimulus, the release of drug was low when compared with the release under electric stimulus ($P < 0.05$). When an electric stimulus was applied, the drug release was greatly enhanced depending on the electric current strength. Under electric stimulus, the drug permeation was greater from ETDDS2 system when compared with ETDDS1 ($P < 0.05$) under identical conditions. This may be attributed to the increased concentration of H-PAAm-*g*-XG copolymer in the hydrogel reservoir, which contains more number of —COO functional groups that lead to increased electrical response and drug release (see Table II). On the other hand, it is interesting to note that in the absence of electric stimulus, the drug permeation from neat SA-based formulation (ETDDS6) was

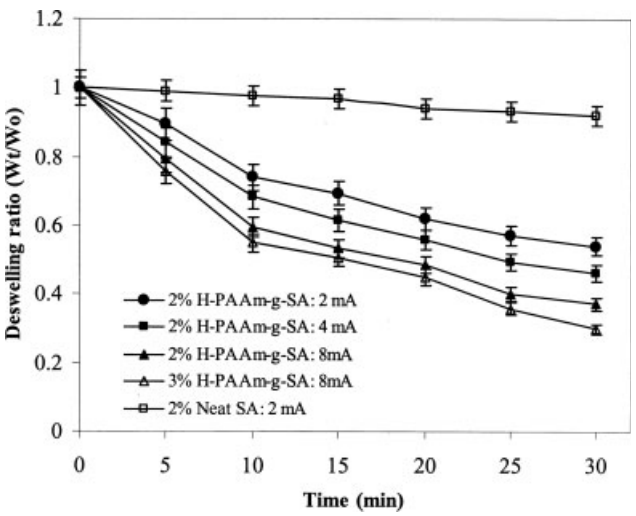

Figure 5 Electrical response of H-PAAm-*g*-SA hydrogel.

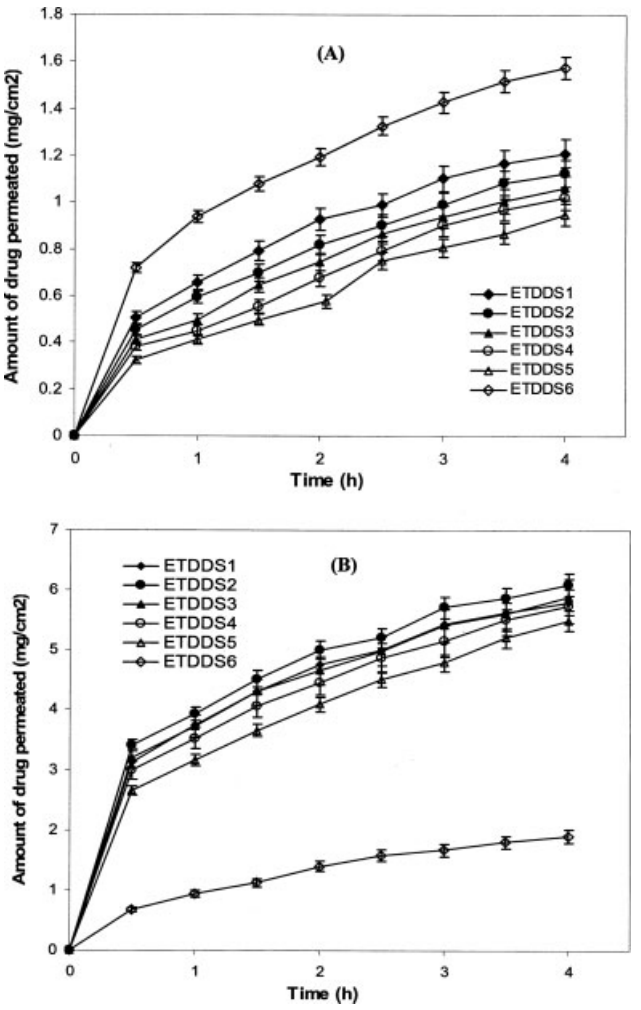

Figure 6 *In vitro* release of ketoprofen from various ETDDS in the absence of electric stimulus (A) and in the presence of 2 mA electric stimulus (B).

higher ($P < 0.05$) when compared with graft copolymer-based formulations, suggesting that release depends on the chain length of copolymer. In the presence of electric stimulus, the drug permeation was higher from graft copolymer-based formulations compared with that in neat SA-based formulation ($P < 0.05$). This may be attributed to the presence of —COO groups on the backbone graft copolymer leading to increased electrical responsiveness and drug release. Figure 6 also demonstrates the effect of thickness and crosslink density of RCM on the ketoprofen release from delivery systems. It was observed that as the concentration of GA was increased in the RCM, the drug release decreased, which may be caused by the increased rigidity of the RCM leading to decreased mesh size and thus it restricts the movement of drug molecules through it. It was also observed that the drug release decreased as thickness of the RCM was increased; it may be due to the increased diffusional path length for drug molecule. Figure 7(A) depicts the effect of electric current strength on drug release. With increasing the current density from 2 to 8 mA, we observed an increase in drug release.

The steady state flux (Jss) was calculated from the slope of the linear portion of the plots drawn between amounts of drug released per unit surface area of skin versus time. The permeability coefficient (K_p) was then calculated using the following equation:

$$K_p = \frac{\text{Jss}}{\text{Cv}}$$

where Cv is the total concentration of the drug in donor compartment. The enhancement factor (EF) was calculated with the following equation:

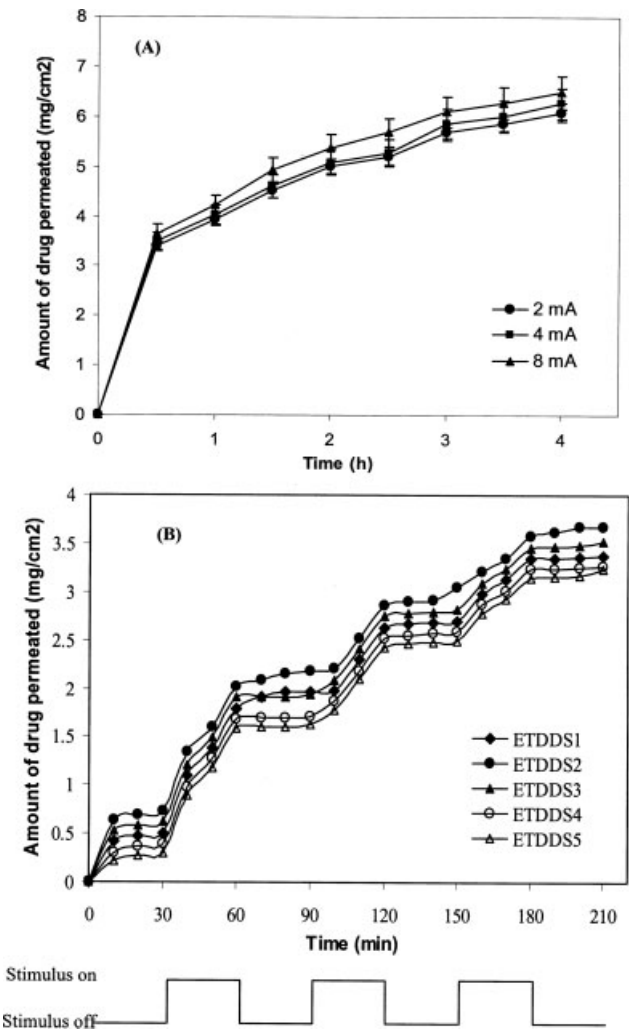

Figure 7 Effect of electric current strength on ketoprofen release from ETDDS2 system (A) and pulsatile release behavior of various ETDDS, in which electric stimulus was switched "on" and "off" at 30 min time intervals (B).

TABLE III
Flux (Jss), Permeability Coefficient (K_p), and Enhancement Factor (EF) of Various Formulations With or Without Electric Stimulus (n = 3)

Formulations	Without electric stimulus		With electric stimulus		EF
	Jss $\times 10^{-2}$ (mg/cm^2/h)	$K_p \times 10^{-2}$ (cm/h)	Jss $\times 10^{-2}$ (mg/cm^2/h)	$K_p \times 10^{-2}$ (cm/h)	
ETDDS 1	26.4	2.6	115.9	11.6	4.4
ETDDS 2	24.7	2.5	120.0	12.0	4.9
ETDDS 3	23.8	2.4	116.2	11.6	4.9
ETDDS 4	23.3	2.3	115.4	11.5	4.9
ETDDS 5	21.6	2.2	112.5	11.2	5.2
ETDDS 6	33.1	3.3	42.7	4.2	1.2

$$\mathrm{EF} = \frac{\text{Jss (with electric stimulus)}}{\text{Jss (without electric stimulus)}}$$

These results are presented in Table III. The flux obtained for all the formulations under passive diffusion ranged between 21.56 $\times 10^{-2}$ and 26.39 $\times 10^{-2}$ mg/cm^2/h, whereas higher values between 112.49 $\times 10^{-2}$ and 120.03 $\times 10^{-2}$ mg/cm^2/h were found under electric stimulus. The applied electric stimulus enhanced the drug release. The obtained EF values ranged between 4.39 and 5.22.

Depending on the electric stimulus turned "on and off," drug release was also turned on and off in a pulsatile manner as shown in Figure 7(B). A rapid release of drug was observed when an electric stimulus was on, which became slower when electric stimulus was off ($P < 0.05$). The possible reason for this type of switching pattern is due to electrically induced changes in the osmotic pressure within the hydrogel matrix. The local pH gradient attributed to electrolysis of the associated water molecules may also change the swelling of polymers as well as the drug release patterns. The drug release varied in proportion to applied intensities of electric stimulus, which gradually increased as the electric stimulus changed from 2 to 8 mA.

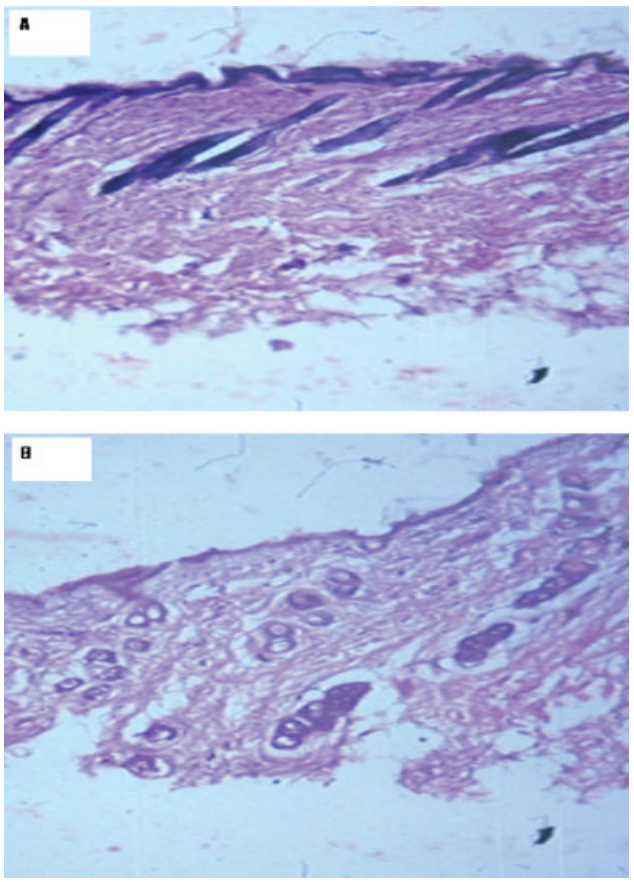

Figure 8 Photographs showing the histopathology of rat skin before application of electrical stimulus (A) and after application of electrical stimulus (B). (Hematoxylin-eosin, 100×). [Color figure can be viewed in the online issue, which is available at www.interscience.wiley.com.]

Histopathological evaluation

The changes in skin structure after subjecting to electrical stimulus are shown in Figure 8 and Table IV. In the case of normal skin (without application of electric stimulus), clearly defined SC could be seen with well-woven structures and there was no inflammatory cell infiltration. A very slight subepidermal oedema and collagen fiber swelling was observed, which may be caused due to the removal of hair from the skin. Skin appendages were found to be normal. On the application of electric stimulus,

TABLE IV
Histopathological Evaluation of Normal Skin and Electrically Stimulated Skin

Parameters	Normal skin	Electrically stimulated skin
Stratum corneum intactness	0	2
Epidermis liquefaction	0	2
Subepidermal oedema	1	2
Collagen fiber swelling	2	3
Inflammatory cell infiltration	0	3
Skin appendages degeneration	0	3

Score: 0, no change; 1, very slight change; 2, slight change; 3, moderate change; and 4, marked change.

the SC was destructed and the cell structure loosened with increased cell infiltration in dermis and degeneration of skin appendages was also observed. This resulted in increased permeation of ketoprofen. The morphological changes in skin structure after application of electric stimulus was also reported earlier.[26]

CONCLUSION

The H-PAAm-*g*-SA hydrogel has undergone deswelling on application of electric stimulus, whereas neat SA did not show any electrical responsiveness. The greater drug release was observed in the presence of electric stimulus, and it was dependent on the applied electric current strength and crosslink density of the RCM. A pulsatile pattern of drug permeation was observed as electric stimulus was switched on and switched off. The H-PAAm-*g*-SA hydrogel used in this study could be useful for the development of transdermal drug delivery systems triggered by electric signal to get on-demand drug release.

The authors thank the financial assistance by Dr. V. Ravichandran Endowment fund for this work.

References

1. Alarcon, C. L. H.; Pennadam, S.; Alexander, C. Chem Soc Rev 2005, 34, 276.
2. Suzuki, H. J Intell Mater Syst Struct 2006, 17, 1091.
3. Murdan, S. J Control Release 2003, 92, 1.
4. Stubble, B. G.; Smedt, S. C.; Demester, J. Pharm Res 2004, 21, 512.
5. Kim, S. Y.; Shin, H. S.; Lee, Y. M.; Jeong, C. N. J Appl Polym Sci 1999, 73, 1675.
6. Peppas, N. A.; Bures, P.; Leobandang, W.; Ichikawa, H. Eur J Pharm Biopharm 2000, 50, 27.
7. Filipcsei, G.; Feher, J.; Zrinyi, M. J Mol Struct 2000, 554, 109.
8. Kulkarni, R. V.; Sa, B. J Appl Biomater Biomech 2007, 5, 125.
9. Agnihotri, S. A.; Kulkarni, R. V.; Mallikarjun, N. N.; Kulkarni, P. V.; Aminabhavi, T. M. J Appl Polym Sci 2005, 96, 301.
10. Hsu, C. S.; Block, L. H. Pharm Res 1996, 13, 1865.
11. Ramanathan, S.; Block, L. H. J Control Release 2001, 70, 109.
12. Kim, S. Y.; Park, S. J.; Kim, I. Y.; Shin, M. S.; Kim, S. I. J Appl Polym Sci 2002, 86, 2285.
13. Kim, S. Y.; Lee, Y. M. J Appl Polym Sci 1999, 74, 1752.
14. Jenson, M.; Hansen, P. B.; Murdan, S.; Frokjaer, S.; Florence, A. T. Eur J Pharm Sci 2000, 15, 139.
15. Kulkarni, R. V.; Sa, B. Curr Drug Del 2008, 5, 256.
16. Kulkarni, R. V.; Sa, B. J Drug Target 2008, 16, 167.
17. Kulkarni, R. V.; Sa, B. Drug Dev Ind Pharm 2008, 34, 1406.
18. Kaur, H.; Chatterji, R. R. Macromolecules 1990, 23, 4868.
19. Kulkarni, R. V.; Sa, B. J Biomater Sci 2009, 20, 235.
20. Palmieri, G. F.; Bonacucina, G.; Di Martino, P.; Martelli, S. J Microencapsul 2002, 19, 111.
21. Eromosele, I. C.; Eromosele, C. O.; Zanna, H. K. J Appl Polym Sci 2002, 84, 500.
22. Mutalik, S.; Udupa, N. J Pharm Pharmaceut Sci 2005, 8, 26.
23. Kulkarni, R. V.; Doddayya, H.; Sagar, P. Pharm Formulation Qual 2007, 9, 39.
24. Pillai, O.; Panchagula, R. J Control Release 2003, 89, 127.
25. Sutani, K.; Kaetsu, I.; Uchida, K. Rad Phy Chem 2001, 61, 49.
26. Bhatia, K. S.; Gao, S.; Freeman, T. P.; Singh, J. J Pharm Sci 1997, 86, 1011.