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RESEARCH PAPER

In Vitro Release Study of Verapamil Hydrochloride Through Sodium Alginate Interpenetrating Monolithic Membranes

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ABSTRACT

Polymeric sodium alginate interpenetrating network membranes containing verapamil hydrochloride were fabricated for transdermal application. The membranes were evaluated for their physical properties, weight and thickness uniformity, water vapor transmission, as well as drug content uniformity. All the thin patches were transparent, smooth, and flexible. The drug-loaded membranes were analyzed by X-ray diffraction to understand the drug polymorphism inside the membrane. The transdermal patches were permeable to water vapor, indicating the permeability characteristics of the polymers. The in vitro drug release was performed in distilled water using a Keshary-Chien diffusion cell. The release data were analyzed to understand the mechanism of drug release.

Key Words: *Interpenetrating network; Sodium alginate; Transdermal matrix; Verapamil hydrochloride*

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INTRODUCTION

Recently, we have been actively involved in studying the controlled release (CR) of antihypertensive drugs using a variety of polymeric matrices (1–5). Apart from the development of oral CR formulations, transdermal drug delivery (TDD) systems using thin polymeric membranes have been widely studied (6–8). The TDDs are precise, consistent, and noninvasive (9). Several transdermal systems containing scopolamine for 72 h prophylaxis of motion-induced nausea (10), nitroglycerine for once-a-day administration in the treatment of angina pectoris, and clonidine (11) for the weekly treatment of hypertension have been suggested. The literature presents many attempts to improve the permeability of the active agents through the skin by various techniques, such as inclusion of keritolytics and penetration enhancers (12,13) to make a more flexible membrane by plasticization (14). The substance added into the matrix may further lead to adverse effects on the skin. Flexibility and plasticization of the membranes can be achieved by blending with another polymer, by cross-linking, or by both cross-linking and blending. Such cross-linked polymeric blends are referred to as interpenetrating polymeric networks (IPNs). The advantages of such systems are not only to create additional free volume space to accommodate the drug without hindrance during its release from the matrix, but also that these systems are biocompatible.

In earlier investigations (3,4,15) from our laboratory, we developed several types of IPNs to study the CR of antihypertensive drugs. Such systems contain two polymers, each in a network form, that can be cross-linked in the presence of each other to give a three-dimensional network structure having large free volume to facilitate the encapsulation of the drug. As a further contribution in this area, we report here the development of IPN films of sodium alginate (Na-Alg) for the release of verapamil hydrochloride (VH), an antihypertensive drug. The fabricated membranes were further characterized by a swelling study to calculate the molar mass between cross-links and thermal analysis of plain and drug-loaded membranes. The in vitro release data performed in distilled water at 37°C for up to 120 min were fitted to an empirical equation to understand the release mechanism and the type of diffusion mechanism. Such release data are important in further development of similar systems by selecting a proper blend mixture.

The drug selected in this research, verapamil hydrochloride, is a calcium channel blocker that has been widely used in the management of hypertension. VH is approximately 90% absorbed by the gastrointestinal tract (GIT), but is subjected to considerable first-pass metabolism in the liver, with a bioavailability of only 20%. The VH exhibits bi- or triphasic elimination kinetics and has a metabolic plasma half-life of about 7 h following a single oral dose or after intravenous administration (11,16). VH acts within 5 min of intravenous administration and in about 1–2 h after oral administration.

EXPERIMENTAL

Materials

A gift sample of verapamil hydrochloride was obtained from Torrent Pharmaceuticals, Ahmedabad, India. Sodium alginate (approximate molecular weight 240,000), guar gum (GG), glutaraldehyde (GA) (25% w/v) solution, and AR grade methanol were all purchased from s.d. Fine Chemicals, Mumbai, India. Water used was double distilled, and its purity was checked by measuring its density (0.9965 g/ml) at 25°C, which agreed well with values in the literature.

Methods

Preparation of Verapamil Hydrochloride-Loaded Transdermal Membranes

The 3% (w/v) Na-Alg and 0.5% (w/v) GG solutions were prepared by dissolving the weighed quantities in distilled water at 40°C on a magnetic stirrer. Then, 60 ml of the prepared 3% Na-Alg solution was put in five conical flasks. We added 9, 18, 27, 36, and 45 ml of 0.5% GG to the flasks and mixed the solutions thoroughly on a magnetic stirrer. After uniform mixing, 35 ml of the polymer blend solution was poured into a glass bangle (5.6 cm diameter) placed on mercury surface in a petri dish (17) and kept in an oven at 40°C for complete drying. Cross-linking was done by dipping the membranes into methanol containing 1% GA and 1% of 1 N HCl for 4 h. The cross-linked membranes were washed in distilled water to remove the unreacted GA. VH was then loaded into the

membrane by soaking the swollen membranes into the VH-containing solution for 1 h. Then, the membranes were dried in air; the rate of evaporation of solvent was controlled by inverting a funnel over the petri dish, which provided constant moisture on the film during drying. After 24 h, the dried patches were taken out and stored in a desiccator for further experiments.

Estimation of Verapamil Hydrochloride in the Membrane

Membranes with a specified area (1 cm^2) were cut into small pieces and put into a 100-ml volumetric flask. About 50 ml of distilled water was added, gently heated to 45°C for 15 min, and kept for 24 h with occasional shaking. Then, the volume was made up to 100 ml with distilled water. Similarly, a blank was carried out using a drug-free patch. The solutions were filtered and centrifuged, and absorbance was measured at a λ_{max} of 229 nm (18) using an ultraviolet (UV) spectrophotometer (Secomam, Anthelie, France).

Permeability Measurement

Water vapor transmission studies were carried out according to the method proposed by Rao and Diwan (19) using glass vials of equal diameter as the transmission cells. These cells were washed and dried in the oven. About 1 g of fused calcium chloride was taken in the cells, and the polymeric patches (1.54 cm^2 area) were fixed over the brim with the help of an adhesive. Then, the cells were accurately weighed and kept in a closed desiccator containing the saturated solution of potassium chloride (200 ml). Humidity inside the desiccator was measured by a hygrometer; the relative humidity (RH) was found to be 84%. The cells were removed and weighed every day for 7 days of storage.

Fourier Transform Infrared Measurements

Fourier transform infrared (FTIR) spectral measurements were performed using a Nicolet model Impact 410 instrument to confirm the chemical interactions between VH and the IPN matrix. The samples (~2 mg) of the pure VH, IPN of Na-Alg, and VH-loaded IPN of Na-Alg IPN were ground with KBr (~148 mg), and pellets were made by applying 6 tons of hydraulic pressure.

Differential Scanning Calorimetry and X-ray Diffraction Studies

Thermal analysis was performed on pure VH, IPN of Na-Alg IPN, and VH-containing IPN of Na-Alg using the DuPont-2000 microcalorimeter (made in the United States). The samples were heated at the rate of 10°C/min under a constant flow of nitrogen gas. The X-ray diffraction (XRD) studies were carried out on the above-mentioned samples using the powder XRD technique with a Philips model PW-1710 diffractometer attached to a digital graphical assembly and computer with Cu-NF 25 KV/20-mA tube as the CuK_α radiation source in the range 0°–90° 2θ (USIC, Shivaji University, Kollapur, India).

In Vitro Drug Permeation Study

In vitro drug release was performed in distilled water using a Keshary-Chien diffusion cell (21). The appropriate size polymeric patches were mounted between the donor and the receptor compartments of the diffusion cell and were held securely by springs. The donor compartment was empty and open to air, but the receptor compartment was filled with distilled water. The magnetic stirrer was set at 100 rpm to circulate water. The whole assembly was maintained at 37°C. The amount of drug released was determined by withdrawing 5-mL aliquots at the selected specific time intervals up to 24 h. The volume withdrawn was replaced with an equal volume of fresh, prewarmed (37°C) distilled water. Samples were analyzed using a UV spectrophotometer (Secomam) at the λ_{max} of 229 nm using distilled water as the blank.

RESULTS AND DISCUSSION

Various blend compositions of Na-Alg with GG were cross-linked by GA to produce IPNs having a three-dimensional structure to facilitate the encapsulation of drugs; the Na-Alg IPN polymers showed good film-forming properties. The films formed were thin (about 40 μm), flexible, smooth, and transparent. The method adopted for casting the films on the mercury surface was quite satisfactory to produce films of uniform thickness. However, one of the requirements of the transdermal membranes is that the films must be compatible with the skin surface. The primary skin irritation test results

conducted on albino rabbit skin per the procedure available (22) indicated that the drug-free transdermal patches produced very slight erythema when compared to Johnson Patch, which was used as a control patch, and no edema was observed for the membrane developed. This indicates that the membranes are skin compatible.

FTIR spectra of the drug, neat polymer, and drug-loaded polymers are presented in Fig. 1. The characteristic FTIR absorption peak of VH (curve a) showed the C–H stretching vibrations of the methoxy group at 2840 cm^{-1}. The N–H stretching of the protonated amine group is observed in the range 2800–2300 cm^{-1}, and a strong absorption band due to C–O stretching of the aromatic ester group appeared at 1262 cm^{-1}. FTIR spectra of the VH-loaded Na-Alg (curve b) show a prominent peak for VH at 1517 cm^{-1}, which is due to benzene rings. This indicates that VH is not involved in any chemical reactions with either the polymer or the cross-linking agent.

DSC scans of the VH-loaded cross-linked Na-Alg matrix (curve a), polymeric matrix (curve b), and pure VH (curve c) are presented in Fig. 2. The endothermic peaks of pure VH appeared at its melting point, 144.5°C (curve c), which did not appear in the DSC plots of the VH-loaded cross-linked Na-Alg matrix (curve a). This further confirms the molecular level dispersion of VH in the polymer matrix. The endothermic peak of the polymeric matrix at 105°C (curve b) after loading the drug decreased to 100°C due to the possible formation of a loose network as the result of the creation of extra free space after drug loading.

The X-ray diffraction patterns of VH (curve a), Na-Alg loaded with VH (curve b), and the

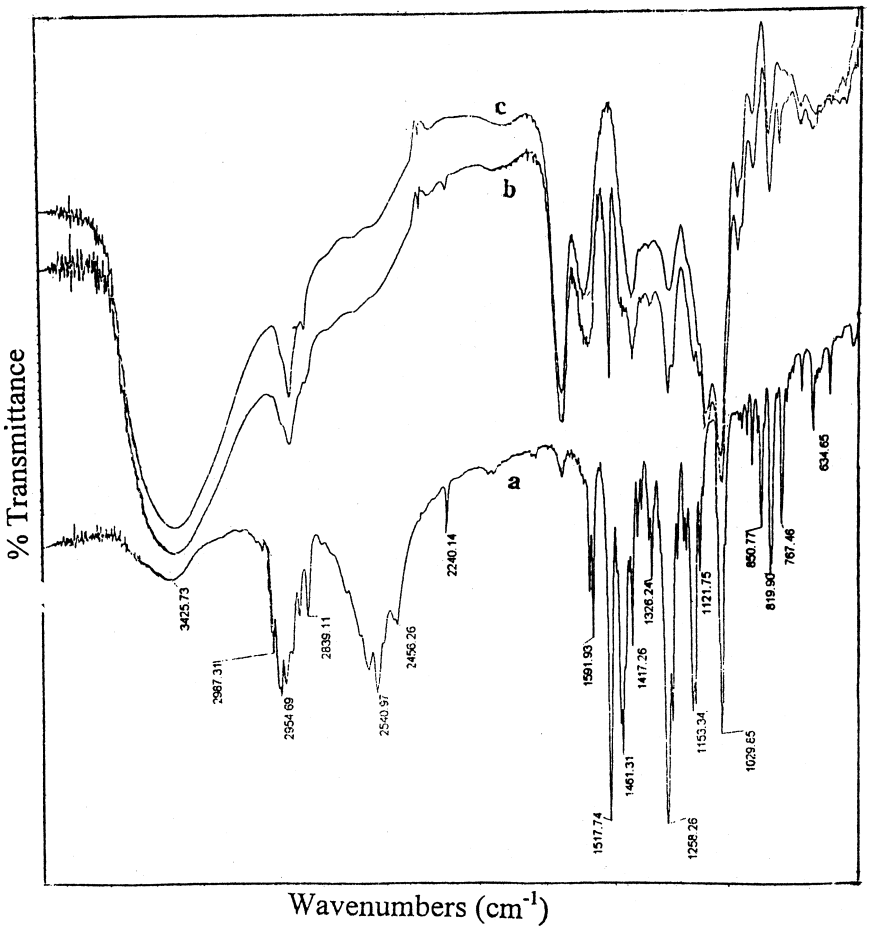

Figure 1. FTIR spectra of (a) VH, (b) VH-loaded Na-Alg IPN, and (c) Na-Alg IPN.

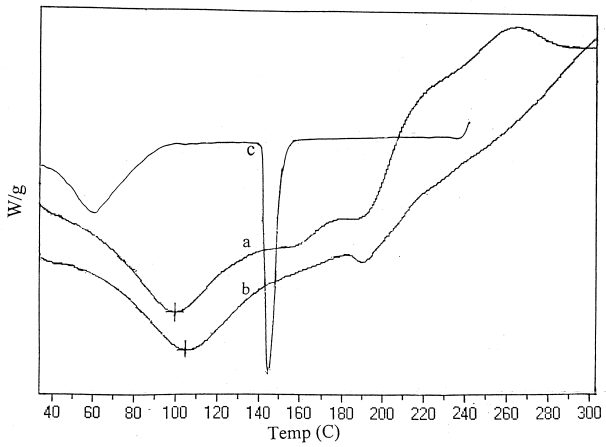

Figure 2. DSC curve of (a) VH, (b) VH-loaded Na-Alg IPN, and (c) Na-Alg IPN.

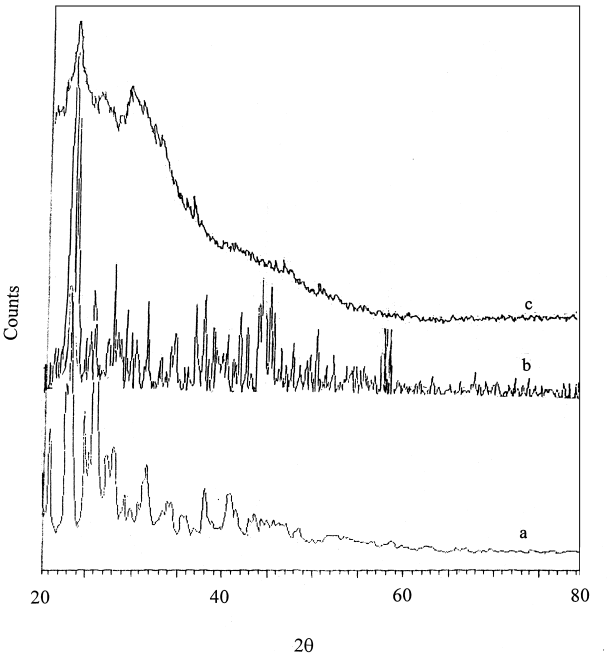

Figure 3. XRD spectra of (a) VH, (b) VH-loaded Na-Alg IPN, and (c) Na-Alg IPN.

cross-linked Na-Alg membrane (curve c) are presented in Fig. 3. The cross-linked polymer shows two types of crystals, one at around 28° and other at 35° (curve c). Unlike the polymeric X-ray diffraction patterns, VH shows many characteristic sharp peaks (curve a), which also appeared in the X-ray diffraction spectra (curve b) of the VH-loaded Na-Alg matrix, but the peak intensities are less when compared to pure drugs. This indicates that the drug loaded inside the polymer matrix is not completely in the crystalline form. This also confirms that a portion of the drug molecules is uniformly dispersed in the polymeric membrane.

The water vapor transmission (WVT) data through the transdermal films are important in knowing the permeation characteristics of the films. The WVT rates were calculated using the following formula (23):

$$\text{Transmission rate} = WL/S \tag{1}$$

where W is the weight of water transmitted in $g \cdot cm^2$/day, L is the thickness of the film, and S is the exposed surface area of the film. The results for the WVT rates are presented in Table 1 and show that all the IPN films were permeable to water vapor and that the WVT data followed zero-order release kinetics. The WVT results are displayed in Fig. 4. Increasing the concentration of GG in the IPN increased the WVT rate, and these values ranged from (2.59–2.80) $\times 10^{-4} g \cdot cm^2$/day.

Swelling of the membrane plays an important role in controlling the drug transport. The membrane swelling in the case of IPN network polymers and the subsequent release of VH from the polymer depends on the extent of cross-linking (5). It is therefore important to study the effect of cross-linking on the percentage loading of VH. To study this effect, the dynamic swelling results were used to compute the molecular mass M_C between the cross-links using the simplified Flory-Rehner equation of Kulkarni et al. (24):

$$M_C = -\rho V \phi^{1/3}[\ln(1-\phi) + \phi + \chi\phi^2]^{-1} \tag{2}$$

Here, the volume fraction ϕ refers to the swollen IPN polymer, and ρ is its density; χ is the interaction parameter, and V is the molar volume of the solvent. The calculated M_C values following the procedure published earlier (25) are presented in Table 1. Since the IPN polymers used in this research are hydrophilic, water transport through these films depends on the rigidity of the polymer backbone in addition to the extent of cross-linking. Thus, the polymer, which had more GG, exhibited reduced swelling of the Na-Alg polymer, but the M_C values tended to increase.

An increase in percentage composition of GG in the IPNs with an increase in VH loadings was observed. This may be due to the fact that swelling of the GG is higher than that of the

Table 1

Results of Water Vapor Transmission (WVT) Rate, Diffusion Coefficients (D) *of the Drug Verapamil Hydrochloride (VH) and* n *Values Calculated from Eq. 3*

GG:Na-Alg Mass Ratio	WVTR ($g \cdot cm^2/day$) $\times 10^4$	VH Content (mg/cm^2)	D (cm^2/s)$\times 10^8$	M_C	Eq. 3		
					k	n	r
0.2 : 8.0	2.59	0.866	2.42	128	0.056	0.51	0.998
0.4 : 8.0	2.80	0.952	2.64	138	0.062	0.51	0.993
0.6 : 8.0	2.85	1.010	3.04	204	0.080	0.48	0.994
0.8 : 8.0	2.88	1.039	3.17	300	0.087	0.46	0.997
1.0 : 8.0	2.93	1.097	3.23	328	0.089	0.49	0.998

GG, guar gum; Na-Alg, sodium alginate.

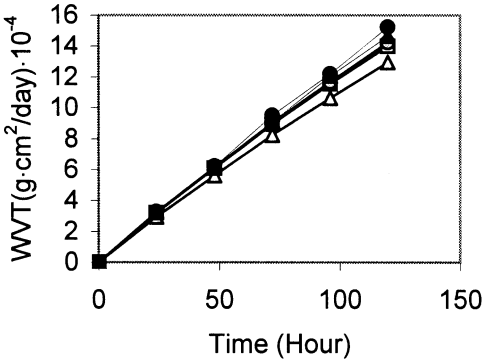

Figure 4. WVT through Na-Alg IPN matrix containing △, 0.2; □, 0.4; ○, 0.6; ▲, 0.8; and ●, 1.0 mass% GG in 8.0 mass% Na-Alg.

Na-Alg matrix, which normally takes about 100 times the water compared to its dry mass (26). Hence, an increase in the amount of GG in the IPN matrix increases membrane swelling. This results in an increase of water content at a higher percentage loading of VH.

The in vitro drug release studies were carried out in distilled water at 37°C using the Keshary-Chien diffusion cell. The membranes developed here were hydrophilic in nature and were converted partly into the hydrophobic type either by cross-linking or by network formation. When these membranes were brought in contact with water, the polymer chain relaxed and swelled. However, in an actual situation (i.e., when applied on the skin), there is insufficient water to relax the polymer chain; therefore, the release data obtained in water at 37°C may not be the perfect condition for in vitro studies. To find the nature of the release kinetics and the mode of drug diffusion, the in vitro release data were obtained for only a few hours (120 min).

The percentage release data displayed in Fig. 5 indicate that an increase in the amount of GG in the IPN increases the release of the drug. This suggests that more free volume is available, possibly due to the formation of an IPN. This is further supported by the higher percentage loading of VH in the IPN. From the plots of cumulative release versus time, the amount of VH released at 18 h was calculated, and these data are presented in Fig. 6. It is observed the release of VH is higher for the IPNs containing more GG. For instance, 559, 738, 766, 766, and 775 μg of VH were released, respectively, when 0.2%, 0.4%, 0.6%, 0.8%, and 1.0% of GG were present in the IPNs. The membranes became porous after the dissolution experiments due to leaching of the drug and the un-cross-linked polymer, as evidenced by the scanning electron micrograph taken of the membranes before and after the dissolution (not shown in figure).

The results of fractional release M_t/M_∞ of VH were analyzed using the empirical equation (27)

$$M_t/M_\infty = kt^n \tag{3}$$

In the Eq. 3, n represents the diffusion anomalies, whereas k is an interaction parameter between the drug and the membrane polymer. Using the least-squares procedure, we estimated the values of n and k; these results, along with the correlation coefficient r values are presented in Table 1. In the case of solute release from the swellable matrices, if $n = 0.5$, then transport follows a Fickian trend (28). For all the systems studied in this work, the values of n

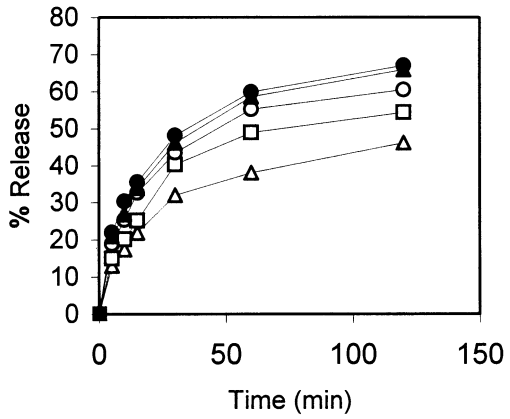

Figure 5. Percentage release of VH from Na-Alg IPN matrix containing △, 0.2; □, 0.4; ○, 0.6; ▲, 0.8; and ●, 1.0 mass% GG in 8.0 mass% Na-Alg.

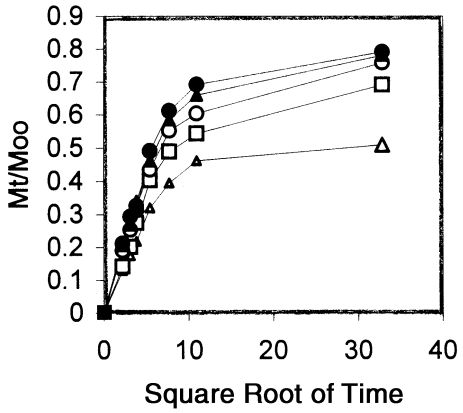

Figure 7. Plot of M_t/M_∞ of VH from Na-Alg IPN matrix containing △, 0.2; □, 0.4; ○, 0.6; ▲, 0.8; and ●, 1.0 mass% GG in 8.0 mass% Na-Alg versus root time (min).

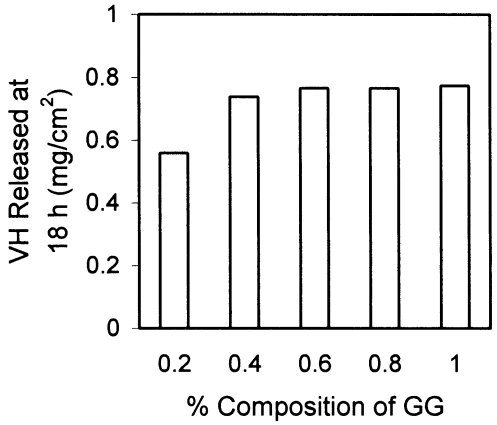

Figure 6. VH released at the 18th hour from Na-Alg IPN.

ranged between 0.46 and 0.51, suggesting that the diffusional transport follows the Fickian model. The plots of M_t/M_∞ versus $t^{1/2}$ for all the IPNs are shown in Fig. 7, which indicates the initial straight-line behavior and later deviations from linearity, indicating that the initial portion of the curve follows a Fickian diffusion (Higuchi release) mechanism, that is, $n = 0.5$.

Further, the translational diffusion coefficient D or VH release was calculated using the equation derived from Fick's second law (29):

$$D = \left(\frac{h\theta}{4M_\infty}\right)^2 \pi \tag{4}$$

where θ is the slope of the linear portion of the plot of M_t/M_∞ versus $t^{1/2}$, h is the thickness of the membrane, and M_∞ is the maximum release. The values of D calculated per the procedure published earlier (24) are also included in Table 1. The D values are in the range 2.42×10^{-8} to 3.23×10^{-8} cm^2/s. These values increase with an increase in GG concentration in the matrix.

CONCLUSIONS

Monolithic transdermal delivery systems of Na-Alg IPNs were developed for the CR of verapamil hydrochloride. The polymeric IPN films developed in this research are capable of loading VH up to 1.097 mg/cm^2. The in vitro drug release as performed in distilled water at 37°C exhibited 559, 738, 766, 766, and 775 μg of VH release, respectively, for the contents of 0.2%, 0.4%, 0.6%, 0.8%, and 1.0% GG in the IPNs at the 18th hour. Release data were fitted to an empirical equation to calculate n values, which ranged between 0.46 and 0.51, suggesting that the diffusion follows the Fickian trend.

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