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In Vitro Release Studies on Matrix Type Transdermal Drug Delivery Systems of Naltrexone and Its Acetyl Prodrug

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ABSTRACT Matrix type acrylic adhesive transdermal patches of naltrexone (NTX) and its 3-O-acetyl ester prodrug were prepared and evaluated for drug content, thickness, and in vitro release characteristics. Among the four DURO-TAK® adhesive polymers (87-2516, 87-2054, 87-2501, and 87-2582) tested, 87-2516 proved to be the most suitable and compatible polymer for the transdermal delivery of NTX from NTX and prodrug patches. A linear relationship was observed for release flux (F) and cumulative amount (M_t) values versus 1%, 2%, and 3% drug loading at equimolar levels. The release of NTX from the patches showed a good correlation ($R^2 > 0.99$) for M_t vs. $\sqrt{t}$ profiles, indicating that a Higuchian matrix diffusion mechanism of drug release from the transdermal adhesive patches was obtained. Overall, the amounts of NTX released from the prodrug patches were significantly higher than from the NTX patches, at all three drug loading levels.

KEYWORDS Naltrexone, Prodrugs, In vitro release testing, Drug in adhesive patches

INTRODUCTION

Naltrexone (NTX) is an orally active and long acting potent narcotic antagonist, and is also approved by the Food and Drug Administration (FDA) for the treatment of alcoholism in formerly dependent alcoholic patients (Terenius, 1998; Volpicelli et al., 1992). NTX hydrochloride is presently available as a 50 mg film-coated oral tablet (ReVia®) in the United States. NTX is rapidly biotransformed into a less active metabolite, 6-β-naltrexol (Cone et al., 1974). Studies have shown that the lowest effective plasma NTX concentration of 2 ng/mL provided an average of 86.5% blockade of 25 mg intravenous (IV) heroin effects. Thus, in sustained or controlled released therapy for opiate antagonist activity, plasma levels of NTX should be kept above 2 ng/mL (Vereby et al., 1976). NTX possesses a low oral bioavailability (5–40%) due to hepatic first pass metabolism after oral administration (Meyer et al., 1984; Wall et al., 1981). Chronic administration via the oral route is fraught with patient compliance problems, and is also associated with gastrointestinal side effects, including nausea, abdominal pain, and vomiting

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(Kranzler et al., 2000). Therefore, transdermal drug delivery systems may be a better choice for NTX therapy, in order to eliminate first pass metabolism, to allow dosage decreases for decreased toxic effects, and to enable better patient compliance. The therapeutic success of a drug applied in a transdermal patch depends mainly on its ability to permeate through the stratum corneum (SC), the main barrier to successful transdermal delivery rates. Since many drugs do not have the ideal physicochemical properties for efficient skin permeation, different approaches, such as prodrugs, chemical enhancers, iontophoresis, etc., have been developed to improve drug skin permeation. Among the different approaches, the prodrug approach, where conversion to the active compound occurs in the skin or after systemic circulation uptake, is very valuable because it does not alter the barrier property of the SC. Unfortunately, alteration of the SC barrier properties can lead to skin irritation.

NTX permeation through the skin does not achieve the required therapeutic plasma levels, due to NTX's non-ideal physicochemical properties (Chen et al., 1986). By esterifying the 3-O-phenolic group of NTX, various NTX prodrugs have been synthesized (Hussain et al., 1987, 1988). These types of NTX prodrugs were first published for use in increasing oral bioavailability and masking the bitter taste of NTX for buccal delivery (Hussain et al., 1987, 1988). Successful prodrugs for transdermal application should enhance the drug permeation rate, allow ready conversion to parent drug in the skin/body, and should have sufficient chemical stability. Previous studies have shown that 3-O-alkyl ester and 3-O-alkyl carbonate prodrugs of NTX can improve human skin permeation, and that skin permeation correlated with the extent of bioconversion and the prodrug physicochemical properties (Pillai et al., 2004; Stinchcomb et al., 2002). Among the prodrugs tested so far, the 3-O-acetyl ester prodrug of NTX has been shown to have the most successful permeation rate through human skin (Stinchcomb et al., 2002). Transdermal delivery systems need to be developed for NTX and its O-acetyl prodrug in order to move towards clinical testing of an optimized prodrug patch. In the interim, it will be important to compare and contrast NTX and prodrug patches in pigs before clinical testing can begin. Hence, the present investigation was aimed at selecting a suitable adhesive for the development of a matrix type transdermal drug delivery system for both NTX and its O-acetyl prodrug. The prepared transdermal patches of three different drug load percentages were evaluated for uniform drug content, uniform thickness, and appropriate in vitro release characteristics.

MATERIALS AND METHODS

Materials

NTX base was purchased from Mallinckrodt Inc (St. Louis, MO). The 3-O-acetyl prodrug was synthesized from NTX free base (Hussain et al., 1987, 1988). Hanks' balanced salts modified powder and sodium bicarbonate were purchased from Sigma Chemical (St. Louis, MO). 4-(2-Hydroxyethyl)-1-piperazineethanesulfonic acid (HEPES), gentamicin sulfate, trifluroacetic acid (TFA), triethylamine (TEA), methanol, and acetonitrile (ACN) were obtained from Fisher Scientific (Fairlawn, NJ). 1-Octane sulfonic acid sodium salt was obtained from ChromTech® (Apple Valley, MN). DURO-TAK® adhesives 87-2516, 2051, 2054, and 2852 were gifts from National Starch and Chemical Company (Bridgewater, NJ, USA).

Synthetic Procedure for the Preparation of NTX-3-O-Acetyl Prodrug

Acetyl chloride (1.3 g, 16.2 mmol) and triethylamine (1.8 g, 17.6 mmol) in dichloromethane (100 mL) were added to NTX (4 g, 11.7 mmoL). After stirring at ambient temperature overnight, the reaction mixture solution was diluted with chloroform (100 mL), washed with brine, dried over potassium carbonate, and concentrated on a rotary evaporator to give the crude product as a brown oil. Purification by silica gel column chromatography (chloroform/methanol, 50/1) afforded 4.27 g (yield 95%) of the title compound as a white solid. An analytical sample was obtained by recrystallization from hexanes/ethyl alcohol. The melting point was 113–115°C. ^{1}HNMR ($CDCl_3$) δ 0.12–0.18 (2H, m), 0.55–0.58 (2H, m), 0.85 (1H, m), 1.55–1.68 (2H, m), 1.90 (1H, m), 2.15 (1H, m), 2.29 (3H, s), 2.35–2.75 (6H, m), 2.95–3.15 (2H, m), 3.20 (1H, m), 4.70 (1H, s), 5.20 (1H, br s), 6.67 (1H, m), 6.84 (1H, m). HRMS: calculated for $C_{22}H_{25}NO_5$ 383.1734, found 383.1742.

Preparation of Patches

Adhesive patches containing different loadings of NTX and its O-acetyl prodrug (1, 2, and 3% w/w) were prepared by using DURO-TAK® adhesives (National Starch and Chemical Company, USA). Appropriate amounts of adhesive and drug were sonicated for 10 min, cast on the release liner (9742 Scotchpak™, 3M, St. Paul, MN) with a wet film applicator (Paul N. Gardner Company Inc., Pompano Beach, FL) set at a 40 mil thickness, and kept at room temperature for one hour and then at 70°C in an oven for 10 min (to remove any residual solvent). The patches were covered with backing membrane (CoTran™ 9722, 3M, St. Paul, MN), cut into appropriate sizes, and stored in a desiccator for further studies.

Drug Content

Drug content in the patches was estimated by placing a 1 cm^2 patch in a 10 mL volumetric flask. Five milliliters of acetonitrile was added to the flask, and the flask contents were sonicated for 10 min to extract the drug from the patches. The volume was made up to 10 mL with acetonitrile and the samples were analyzed by HPLC for NTX and O-acetyl prodrug concentrations.

Thickness of the Patches

The thickness of the patches was measured with a micrometer. The thickness of the matrix was calculated by subtracting the combined thickness of the backing membrane and release liner from the thickness of the whole patch.

In Vitro Drug Release Studies

A USP Dissolution Apparatus 5 (paddle over disk) (Vankel, Cary, NC, USA) was used to characterize drug release from the patches. A 9 cm^2 patch with the matrix facing up was mounted on a Transdermal Sandwich™ (Hanson Research Corp., Chatsworth, CA). The release studies were performed in 900 mL of isotonic phosphate buffer, pH 7.4 at 32°C at a rotation speed of 50 rpm. Samples (5 mL) were collected at different time intervals for 48 hrs. Volumes of fresh buffer (5 mL) were replaced at each sampling time point to keep the volume constant.

Quantitative Analysis

A modification of the high-pressure liquid chromatography (HPLC) assay described by Hussain et al. (1987) was used for the analysis of NTX and its O-acetyl prodrug. The HPLC system consisted of a Waters 717 plus Autosampler, Waters 1525 Binary HPLC Pump, and Waters 2487 Dual Wavelength Absorbance Detector with Breeze software. A Brownlee C-18 reversed-phase Spheri-5 μm column (220 × 4.6 mm) with a C-18 reversed phase 7 μm guard column (15 × 3.2 mm) was used with the UV detector set at a wavelength of 215 nm. The mobile phase was comprised of 70:30 ACN and 0.1% TFA with 0.065% 1-octane sulfonic acid, sodium salt, and adjusted to pH 3.0 with triethylamine. The flow rate of the mobile phase was 1.5 mL/min and 100 μL of sample was injected onto the column. Dissolution samples were analyzed with a set of standard samples and exhibited excellent linearity over the entire concentration range employed in the assays.

The drugs were extracted from the buffer samples by solid phase extraction (30 mg 1 cc Oasis® HLB, Waters Corp., Milford, MA). Before loading the aqueous drug samples (5 mL), the cartridge was conditioned with 1 mL of methanol and 1 mL of water. After loading the sample, the cartridge was washed with 1 mL of 5% methanol and the drug was eluted with ACN and analyzed by HPLC.

Data Analysis

The fraction of the drug released into the dissolution medium (M_t/M_∞) from different adhesive patches can be calculated from the modified Higuchi Eq. 1:

$$\frac{M_t}{M_\infty} = \sqrt{\frac{4D_p t}{L^2 \pi}} = k\sqrt{t} \quad 0 \leq M_t/M_\infty < 0.6 \qquad (1)$$

where M_t is the cumulative amount of drug released at time t, M_∞ is the drug loaded in the adhesive patch, D_p is the apparent diffusion coefficient of drug in the adhesive patch, k is the release rate constant (time$^{1/2}$), and L is the thickness of the adhesive film.

Eq. 1 can be further simplified to:

$$M_t = Ft^{1/2} \qquad (2)$$

where M_t is cumulative amount of drug released per unit area at a time t and F is the release flux derived

TABLE 1 Drug Contents and Thicknesses of Different Adhesive Patches

Patch	Drug content (mg/cm^2)[a]	Thickness (μm)[b]
NTX 1%-87-2516	0.533±0.012 (2.34)	197±6.74 (3.42)
NTX 1%-87-2054	0.532±0.021 (3.94)	202±7.52 (3.72)
NTX 1%-87-2051	0.591±0.01 (2.01)	216±10.30 (4.76)
NTX 1%-87-2852	0.554±0.005 (2.01)	216±7.5 (3.47)
NTX 2%-87-2516	1.220±0.01 (0.81)	204±9.66 (4.73)
NTX 3%-87-2516	1.683±0.06 (3.56)	208±4.16 (2.0)
NTX-Acetyl 1%-87-2516	0.626±0.012 (1.91)	208±9.18 (4.41)
NTX-Acetyl 2%-87-2516	1.477±0.05 (3.99)	221±9.44 (4.49)
NTX-Acetyl 3%-87-2516	2.095±0.075 (3.59)	226±10.05 (4.46)

Values in parentheses are % CV.
[a] $n=3$.
[b] $n=10$.

from the slope of the M_t vs. $t^{1/2}$ profile. The statistical analysis of the data was computed with a one-way ANOVA using SigmaStat software (SPSS, Inc., Chicago, IL) at a significance level of $p<0.05$.

RESULTS AND DISCUSSION

Drug Content and Thickness of the Patches

The drug content and thickness of the patches are given in Table 1. The NTX patches were pale yellow in color and those of the acetyl prodrug were colorless. Both patches had a smooth surface. Small standard deviations (SD) and coefficients of variation (CV) were observed for drug content and thickness data, indicating uniformity in drug content and patch thickness.

In Vitro Drug Release Studies

Choosing an ideal adhesive is important in designing a matrix type adhesive transdermal drug delivery system, since adhesion, chemical stability, and compatibility with other components of the system severely impact the release properties of the drug candidate. In vitro release tests are not only useful in quality control testing of the finished adhesive transdermal drug delivery system, but are also useful for selecting suitable adhesives for formulation of the

TABLE 2 Chemistry and Solvent Composition of Selected Adhesives in the Study

Adhesive	Chemical composition	Functional groups	Cross linker	Solvent composition	Tack (Oz/in^2)
87-2516	Acrylate-Vinyl acetate	-OH	Present	Ethyl Acetate (63%), Heptane (8%), Ethanol (27%), Methanol (2%)	30
87-2051	Acrylate-Vinyl acetate	-COOH	Not present	Ethyl Acetate (87%), Heptane (13%),	80
87-2054	Acrylate-Vinyl acetate	-COOH	Present	Ethyl Acetate (36%), Isopropanol (36%), Heptane (24%), Toluene (4%), 2,4-Pentanedione (<1%)	45
87-2852	Acrylate	-COOH	Present	Ethyl Acetate (65%), Isopropanol (19%), Hexane (12%), Toluene (2%), 2,4-Pentanedione (<1%)	40

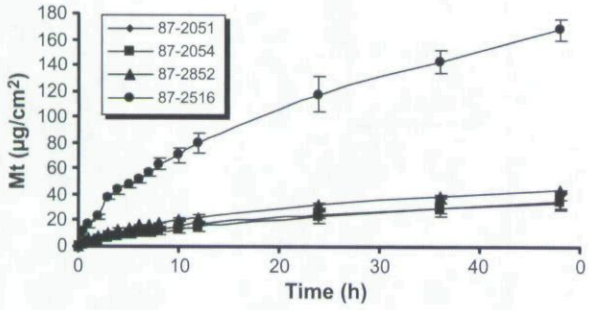

FIGURE 1 Cumulative Release Profiles of NTX from Different Adhesives with 1% Loading (n=3).

matrix type patches. In order to select a suitable adhesive for the NTX and acetyl prodrug patch development, various acrylic adhesives with either hydroxyl or carboxylate functional groups, and with or without cross linking agents, were selected as listed in Table 2. These adhesives were selected based on their chemical composition, solvent compatibility, and tack properties, in order to design an ideal patch for NTX and its prodrug. Initial studies were carried out by loading 1% w/w NTX in different adhesives, and the drug release properties were evaluated from these release studies.

The cumulative release profiles of NTX from the four different adhesives are shown in Fig. 1. A steady and continuous release of NTX from all the patches was observed, indicating that NTX was dissolved in all the adhesives with a uniform distribution. In all of the cases, the release kinetics followed a square root of time relationship. The profiles of the fraction of the drug released into the dissolution medium (M_t/M_∞) from different adhesives with 1% NTX loading vs. $\sqrt{t}$ are shown in Fig. 2. The release rate constant (k) and apparent diffusion coefficient (D_p) values calculated from Eq. 1 for NTX from different

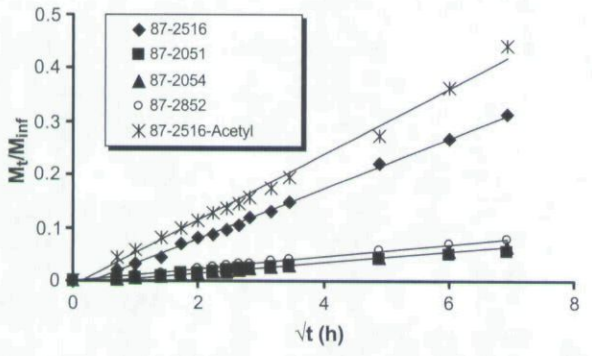

FIGURE 2 Release Kinetics of NTX from Different Adhesives at 1% Loading.

TABLE 3 Release Rate Constant (k) and Apparent Diffusion Coefficient (D_p) Values of NTX from Different Adhesives and Prodrug from 87-2516 Adhesive at 1% Loadings

Adhesive	k ($h^{1/2}$)	D_p (cm^2/h)
87-2516	0.039	4.66×10^{-7}
87-2051	0.008	2.42×10^{-8}
87-2054	0.009	2.45×10^{-8}
87-2852	0.011	4.79×10^{-8}
87-2516 prodrug	0.059	1.19×10^{-6}

adhesives are given in Table 3. The release rate of NTX from the 87-2516 adhesive was 4.8, 4.5, and 3.4 times higher than the 87-2051, 87-2054, and 87-2852 adhesives, respectively (1% NTX loading). Overall, the order of the release rates from all the adhesives was 87-2516>87-2582>87-2054=87-2051. A similar ranking for the apparent diffusion coefficient (D_p) values for NTX in the different adhesives was also observed (Table 3). These results indicated that 87-2516 is a suitable and compatible polymer for the development of transdermal drug delivery systems of NTX and its O-acetyl prodrug.

Further studies were carried out by preparing 87-2516 adhesive patches with different loadings of NTX and its O-acetyl prodrug. A maximum of 3% NTX can

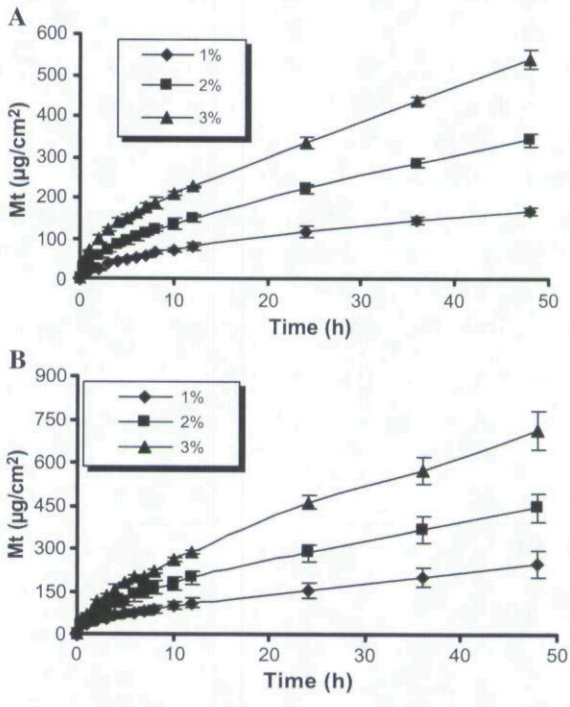

FIGURE 3 Cumulative Release Profiles of NTX from NTX (A) and O-Acetyl Prodrug (B) 87-2516 Adhesive Patches with Different Loadings (n=3).

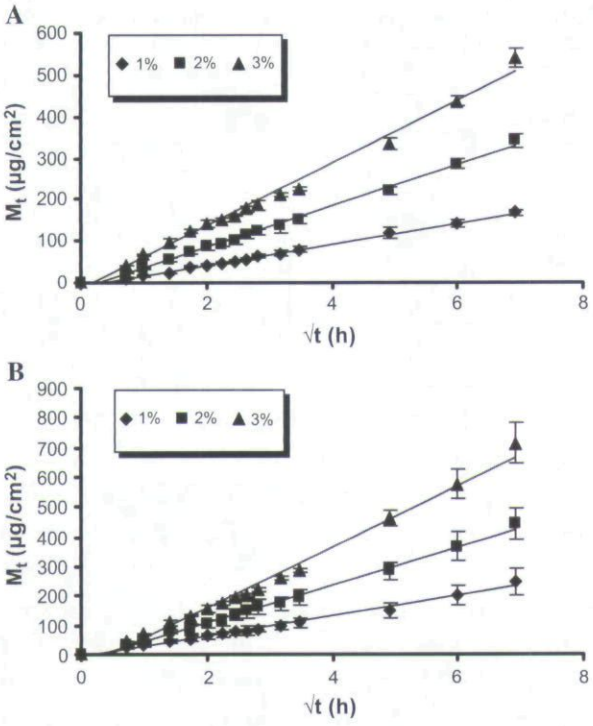

FIGURE 4 Cumulative Amount Released Versus Square Root Time Profiles of NTX from NTX (A) and O-Acetyl Prodrug (B) 87-2516 Adhesive Patches with Different Loadings.

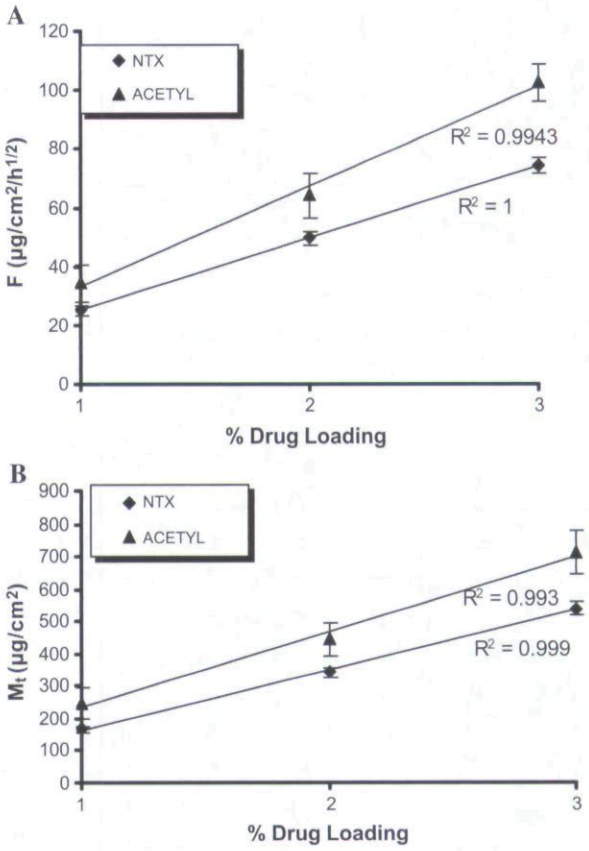

FIGURE 5 Linear Relationships Between % Drug Loading and F (A), M_t (B) Values of NTX from NTX and O-Acetyl Prodrug 87-2516 Adhesive Patches.

be loaded into the adhesive matrix. Above the 3% level, crystallization of NTX was observed. Therefore, 3% levels of both drugs were loaded and the results were compared. The cumulative amount (M_t) released vs. time profiles of NTX from NTX and the O-acetyl prodrug patches with different loadings are shown in Fig. 3A and B. The release flux (F) values obtained from M_t vs. $\sqrt{t}$ profiles (Fig. 4A and B) for NTX from NTX and its O-acetyl prodrug patches are given in Table 4, along with the cumulative amounts of NTX (M_t) released at the end of 48 h. Good correlation coefficients for F values were obtained with M_t vs. $\sqrt{t}$ profiles, indicating that a Higuchian matrix diffusion mechanism of drug release from the transdermal adhesive patches was obtained. The results demonstrated that an increased loading of NTX and its O-acetyl prodrug significantly increased both the F (µg/cm^2/h$^{1/2}$) and M_t values for NTX from these patches ($p<0.05$), with the exception of the F values at 1% loading. A linear relationship was observed for release flux (F) and cumulative amount (M_t) values vs. 1, 2, and 3% drug loading at equimolar levels (Fig. 5). The prodrug patches had consistently higher F and M_t

TABLE 4 Release Flux (F) and Cumulative Amount (M_t) at 48 h for 87-2516 Adhesive Patches with Different Loadings of NTX and Its O-Acetyl Prodrug

Patch	F (µg/cm^2/h$^{1/2}$)[a]	R^2	M_t at 48 h (µg/cm^2)[a]
NTX 1%	25.01±1.82	0.9964	167.84±8.12
NTX 2%	49.44±2.35	0.9952	342.28±16.50
NTX 3%	74.00±2.70	0.9999	539.27±22.52
Prodrug 1%	34.38±6.29*	0.9999	246.86±46.47*
Prodrug 2%	64.09±7.53*	0.9947	445.51±51.02*
Prodrug 3%	102.75±6.29*	0.9872	712±67.35*

[a]$n=3$.
*Values include prodrug and NTX formed in dissolution medium in NTX equivalents.

values on the order of 30–40% above NTX patch values. The increase in F and M_t values for NTX from the O-acetyl prodrug patch over the NTX patch can be very well explained by the higher D_p value observed with the O-acetyl prodrug patch (Table 3).

CONCLUSIONS

The prepared adhesive transdermal patches of NTX and its O-acetyl prodrug showed good uniformity with regard to drug content and thickness. The 87-2516 adhesive polymer was found to be most suitable for the transdermal delivery of NTX from the NTX and O-acetyl prodrug patches. Increases in drug loading linearly increased the F and M_t values of NTX from the NTX and the O-acetyl prodrug patches. Overall, the results of the present investigation demonstrated that significantly increased amounts of NTX can be delivered from 87-2516 transdermal patches with the NTX-3-O-acetyl ester prodrug. Further studies on the in vitro human skin permeation of NTX from these adhesive patches is currently under investigation.

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