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In Vitro Skin Permeation Enhancement of Sumatriptan by Microneedle Application

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Abstract: Different dimensions of commercially available microneedle devices, namely, Admin-Patch® microneedle arrays (MN) (0.6, 0.9, 1.2 and 1.5 mm lengths) and Dermaroller® microneedle rollers (DR) (0.5 and 1mm lengths) were evaluated for their relative efficiency in enhancement of transdermal permeation of Sumatriptan (SMT). Solubility assessment of SMT was carried out using propylene glycol (PG), polyethylene glycol (PEG) in combination with saline (S) at different ratios and the order of solubility was found to be 70:30 > 80:20 > 90:10 %v/v in both PG:S and PEG:S. *In vitro* skin permeation studies were performed using PG:S (70:30 %v/v) as donor vehicle. A significant increase in cumulative amount of SMT permeated, steady state flux, permeability coefficient and diffusion coefficient values were observed after microneedle treatment, and the values were in the order of 1.5mm MN >1.2mm MN >0.9mm MN >1mm DR >0.6mm MN >0.5mm DR > passive permeation. Lag times were significantly shorter after longer microneedle application (0.24h for 1.5mm MN). Arrays were found to be superior to rollers with similar microneedle lengths in enhancing SMT permeation and may be attributed to higher density of microneedles and force of application onto skin. The *in vitro* flux values revealed that 2.5cm^2 area patch is sufficient for effective therapy after treatment of skin with 1.5mm MN. It may be inferred that microneedle application significantly enhances the transdermal penetration of SMT and that it may be feasible to deliver clinically relevant therapeutic levels of SMT using microneedle assisted transdermal delivery systems.

Keywords: Microneedle arrays, Microneedle rollers, Permeation, RP-HPLC, Sumatriptan, Transdermal delivery.

INTRODUCTION

Migraine is a common chronic pain disorder with episodic attacks having an adverse influence on the overall quality of life of migraineurs affecting approximately 15% of the population worldwide or 1 billion people with 19% women and 11% men [1-3]. It is a primary headache characterized by recurrent episodes of headache of moderate to severe pain, localized to one cerebral hemisphere, pulsing, nausea and sensitivity to external stimuli such as light, sound, touch, heat or smell [4-6].

Existing anti-migraine therapy employs potent serotonin 5-HT$_1$ receptor agonists, triptans as the first line drugs for the safe and effective treatment of acute migraine for patients who do not respond to COX-inhibitors [6-8]. Sumatriptan (SMT) is the most widely prescribed drug among them and effective for relieving pain and nausea in 75% of the patients [9-11].

SMT can be administered via oral, intranasal or by subcutaneous injection with an absolute bioavailability of around 14%, 15%, and 96% respectively in these routes and recurrence occurs with all the three routes [7, 12]. On oral administration, migraineurs face hindered gastric emptying with more variability of absorption due to gastric stasis during the attack [13, 14]. Consequently unanticipated and erratic absorption with varying magnitudes of therapeutic effect are seen during the attack [15-17].

With an elimination half-life of only 2 hr, the active drug is eliminated within 4-6 hr in most of the patients. Thus, an optimal product would seek to provide the benefits of rapid and systemic delivery of SMT without the need for an injection, and an extended duration of action exceeding the time course of the attack with an intention of preventing recurrent headaches of migraine [6]. Considering the low bioavailability after oral and intranasal administration, due to presystemic metabolism and incomplete absorption, in addition to the patient non-compliance associated with parenteral administration, an optimal product and route of administration that would provide the advantages of rapid, systemic SMT delivery as obtained with parenteral administration (without the need for an injection) and with a consistent duration of action which exceeds the time course of the patients migraine is of interest [18, 19].

SMT is a hydrophilic drug with a log P value of 0.67 which makes it difficult to permeate through the lipophilic barrier of skin and hence optimizing the transdermal delivery of SMT with suitable permeation enhancement technique is necessary [20]. Some of the works were reported on the

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transdermal permeation of SMT using chemical enhancers, physical enhancement techniques like iontophoresis and two layered dissolving microneedle patches [6, 12, 21, 22]. Use of chemical penetration enhancers was not supposed to be much effective for transdermal delivery in case of hydrophilic molecules [23, 24]. Wang *et al*., also discussed about the skin irritation instigated by iontophoresis at high electric voltage [25]. Literature survey emphasizes that microneedle application can effectively enhance the permeation of macromolecules and several hydrophilic drugs via transdermal route [26-30].

Hence, in the present investigation an attempt was made to deliver SMT *via* transdermal route with microneedle treatment. As well-known now, microneedles are minimally invasive systems which can overcome the barrier of the stratum corneum and help improve the permeation of drug molecules. Commercially available microneedles, namely Dermaroller® microneedle rollers (DR) of lengths 0.5 and 1 mm and AdminPatch® microneedle arrays (MN) of lengths 0.6, 0.9, 1.2 and 1.5 mm, made of stainless steel were selected and evaluated with regard to their efficiency on skin perforation and permeation enhancement of SMT through pig ear skin. The drug permeation in skin is known to depend on a number of factors, e.g., force that is imposed on the microneedles, dimensions and distributions of the microneedles in a device, etc [31, 32]. For this reason, a range of well-defined dimensions and geometry of commercial microneedles have been chosen in this study so as to determine how significant the changes (e.g., improvement) in permeability are as these dimensional parameters are changed.

MATERIALS AND METHODS

Materials

AdminPatch® microneedle arrays were purchased from AdminMed, Sunnyvale, U.S.A and Dermaroller® microneedle rollers from DermaIndia, Chennai, India. Sumatriptan Succinate was purchased from Aurabindo pharma Ltd, Hyderabad, India; Sodium chloride, isopropyl alcohol, propylene glycol from LobaChemie, Mumbai, India; Ammonium acetate, acetonitrile, methanol and HPLC water from Merck Specialities Pvt. Ltd, Mumbai, India; Haematoxylin and eosin stain from Sigma-Aldrich, India. All the reagents and chemicals used in the study were of HPLC grade. Pig ear skin was obtained from Local abattoirs.

Methods

Analysis of Samples

A validated RP-HPLC-PDA method was used for the analysis of SMT in the collected samples [33]. A Shimadzu Prominence HPLC system provided with DGU-20A3 degasser, LC-20AD binary pumps, SIL-20AHT auto sampler and SPD-M20A PDA detector was used. Data acquisition was carried out using LC solutions software. A Phenomenex C_{18} reverse-phase column (150 × 4.6 mm; 5 µ) was used with the PDA detector set at a wavelength of 230 nm to perform the chromatographic analysis. The mobile phase comprised of 10 mM ammonium acetate - methanol 75:25 (v/v). Flow rate of the mobile phase was maintained at 1.2 mL/min with an injection volume of 10 µL. The retention time for SMT was 3.61 min and showed a good linearity in a concentration range of 0.2 to 3.0 µg/ml with a correlation coefficient greater than 0.999.

Solubility Studies of SMT

The solubility studies of SMT were carried out in different vehicle systems of propylene glycol (PG), polyethylene glycol (PEG 400), and saline combinations at 70:30% v/v, 80:20% v/v, and 90:10% v/v ratios. To each vehicle system, excess amount of SMT was added and vortexed for 1 min in order to obtain a saturated solution and the solutions were equilibrated at 32°C in an orbital shaker for 24 h. After equilibration, the samples were centrifuged at 3000 rpm for 15min and filtered through a nylon syringe filter (0.45µm) and all the samples were analyzed by RP-HPLC-PDA method after appropriately diluting with saline.

Skin Preparation

Pig ears were collected from the local abattoirs immediately after animals were killed by electric current. The ears were transported to the laboratory in a cooling box without previous treatment. Freezing of the skin was avoided during transport. In the laboratory, the pig ears were washed carefully with distilled water. The hair was removed from the external part of pig ear skin using an electrical hair clipper. Then, carefully the full-thickness skin from the external part of the pig ear was separated from underlying cartilage using a scalpel and excess fat underlying the skin was removed. Pig ear skin of a thickness of 1.2 mm was employed for the *in vitro* transdermal permeation studies. Dermis side was wiped with isopropyl alcohol to remove the residual adhering fat. Processed skin samples were individually wrapped in plastic bags without air entrapment and stored in a deep freezer at -20°C till further use. The pig ears were collected and processed as per the protocol approved by the Institutional Animal Ethics Committee (IAEC).

Skin Penetration by Microneedles

Prior to the skin permeation experiments, the skin samples were taken from the freezer and brought to room temperature for about 30min. After thawing, skin surface was carefully washed with saline. DR (0.5 and 1mm of conical shaped needles) and MN (0.6, 0.9, 1.2 and 1.5 mm having needles of rectangular cross sectional shape in a plane parallel to the substrate) were utilized to poke over the skin surface for a single time in one direction. The microneedles were periodically checked in between usage for potential damages of the needles under a stereomicroscope. The DR and MN were presented in (Fig. **1**, **2**).

Histological Examination of Skin Samples with and without Microneedle Treatment

The histological sections of the skin samples with and without microneedle treatment (negative control) were prepared and observed under a Biological microscope (Olympus; Noida, India). Skin samples were stained with haematoxylin and eosin for visualization of skin layers and to display a clear indentation by microneedle penetration. The depth of microneedle penetration was also calculated with the help of Toup View 3.2 Software (Irwin, U.S.A).

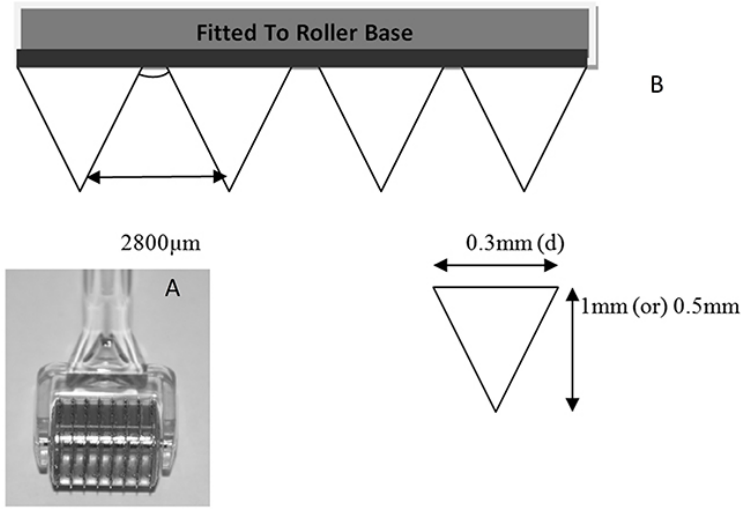

Fig. (1). (**A**) Dermaroller® microneedle roller; (**B**) schematic representation of a microneedle orientation.

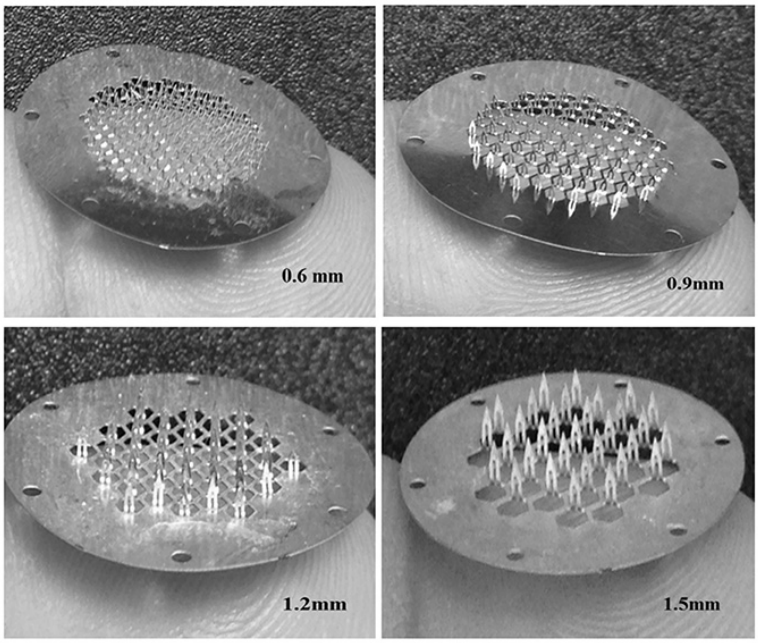

Fig. (2). Adminpatch® microneedle arrays used in the study.

In Vitro Skin Permeation Studies

The *in vitro* transdermal permeation studies were performed using a vertical type Franz diffusion cell apparatus with a water circulation system, a water heater and an eight stage magnetic stirrer (Orchid Scientifics, Nasik, India). Franz diffusion cells with an effective diffusion area of 1.77 cm^2 and a receptor volume of around 14 mL were used. Saline was used as the receptor fluid. Pig ear skin was mounted between donor and receptor cells with stratum corneum surface facing towards the donor cell and clamped. The receptor medium was magnetically stirred for uniform drug distribution at a speed of 600 rpm. Care was taken to prevent any air bubbles between the underside of the skin (dermis) and receptor solution. The surface of the skin was maintained at 32°C using a circulating water bath. After equilibration for 30 min, 500 µL of donor solutions (PEG:S and PG:S combinations) containing excess amount of SMT were applied on to the skin. Samples were withdrawn from the receptor fluid (500 µL) at six hour increments up to 48 hr and replaced with the same volume of fresh saline to maintain a constant volume. All the samples were stored at 4°C, until analyzed by HPLC.

Evaluation of Skin Permeation Data

The cumulative permeation profiles were plotted for cumulative amount of drug permeated (nmoL/cm^2) as a function of time, for untreated and microneedle treated skin. The flux values and the respective lag times were obtained from the slope and the X-intercept of the steady state portion of the cumulative permeation profiles. Apparent permeability

and diffusion coefficient values were computed from Fick's first law of diffusion:

$$\frac{1}{A}\left(\frac{dM}{dt}\right) = J_s = K_p \Delta C$$

J_s is the steady-state flux (nmoL/cm²/hr), M is the cumulative amount of drug permeating the skin (nmoL/cm²), A is the area of the skin (1.77 cm^2), K_p is the apparent permeability coefficient (cm/hr), and ΔC is the difference in concentrations of SMT in the donor and receiver. Sink conditions were maintained in the receiver throughout the experiment and hence ΔC was approximated to the drug concentration in the donor compartment.

SMT Content in Skin

After the completion of permeation studies, skin samples were studied for drug disposition. The skin tissue exposed to the donor solution was expurgated with a scalpel; washed with filtered water and blotted with a paper towel in order to remove the drug adhered to the surface. The skin was minced with a scalpel, and placed in a pre-weighed vial. The drug was extracted from the skin by equilibrating with 1 mL of acetonitrile at 32°C with shaking (100 rpm) on a vortex shaker. Processed skin samples were analyzed by HPLC to determine the SMT content.

Statistical Analysis of the Data

Results of the experimental data were subjected to statistical analysis by one way ANOVA using SYSTAT 13 software (Systat Software Inc., San Jose, USA). Results with *p* value of less than 0.05 were considered to have statistically significant variance. Mean of replicate measurements (n = 3) with corresponding standard deviation (SD) was used to represent the data and to plot the graphs.

RESULTS

Solubility Studies of Sumatriptan

Solubility studies of SMT were performed in different vehicle combinations of PG:S and PEG:S (70:30, 80:20 and 90:10 %v/v) with a view to select appropriate donor vehicle of SMT for *in vitro* skin permeation studies. The solubility data obtained was illustrated in (Fig. **3**). From the data obtained, PG:S combinations (70:30, 80:20 and 90:10 %v/v) and PEG:S (70:30 %v/v) were selected for further *in vitro* permeation studies.

Histological Examination of Skin Samples with and without Microneedle Treatment

Histological sections were prepared using haematoxylin and eosin stain (H&E stain) and observed with biological microscope (Olympus; Noida, India) under a magnification of 40x and were presented in the (Fig. **4**). From the figures the pokes and stratum corneum disruption at poked sites were clearly evident. Depth of penetration was calculated using Toup view software and was found to be approximately 30-40% for DRs and 40-50% for MNs of their respective needle lengths.

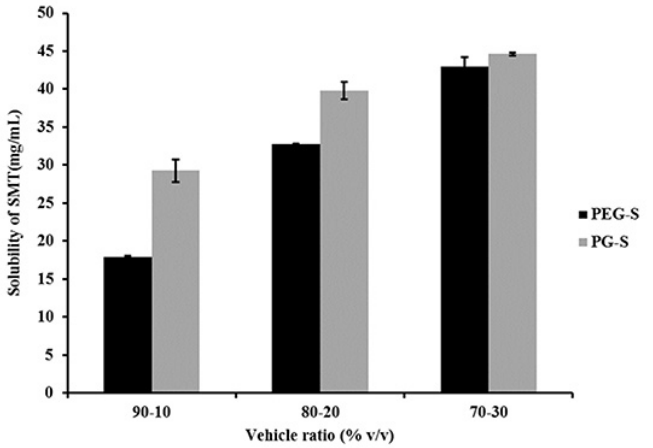

Fig. (3). Solubility of SMT in PG-S and PEG-S combinations.

In-vitro Skin Permeation Studies

Passive Permeation

Initially, passive permeation studies (without microneedle treatment) were carried out using the selected donor vehicles. Cumulative amounts of SMT permeated at the end of 48hr for PG-S (70:30, 80:20 and 90:10 %v/v) were 11375.40±1455.59, 5677.85±879.91 and 4432.61±874.43 nmoL/cm² respectively and for PEG-S 70:30 %v/v was 9241.96±130.34 nmoL/cm². Comparative *in vitro* passive permeation profile of SMT was shown in the (Fig. **5**).

Similarly, the steady state flux values obtained for PG-S vehicle systems at 70:30, 80:20, and 90:10 %v/v compositions were 255.80±31.82, 122.54±18.15 and 86.10±19.11 nmoL/cm^2/hr respectively and that for PEG-S 70:30 %v/v was 190.25±3.11 nmoL/cm^2/hr. The results revealed that the flux value obtained was higher for PG-S at 70:30 %v/v similar to the cumulative amount of SMT permeated.

Microneedle Pre-Treatment to the Skin

After microneedle treatment of the skin, the permeation studies were carried out using PG:S 70:30 %v/v as donor vehicle. The cumulative amounts of SMT permeated at the end of 48 hr were found to be 40485.54±3977.06, 184080.37 ±9450.87, 275904.49±5005.44 and 325491.12±26393.48 nmoL/cm^2 respectively for 0.6, 0.9, 1.2 and 1.5 mm MN and were 21027.88±3673.23 and 150433.28±28569.25 nmoL/cm^2 respectively for 0.5 and 1 mm DR. Comparative *in vitro* permeation profiles were plotted as a function of time for passive and microneedle treatment permeation studies (Fig. **6**).

Furthermore, microneedle treated steady state fluxes were 895.93±83.13, 3725.14±346.28, 5636.61±130.87 and 6754.24±615.94 nmoL/cm^2/h respectively after treatment of the skin with 0.6, 0.9, 1.2 and 1.5 mm MNs and 513.27± 94.67 and 3302.9±496.63 nmoL/cm^2/h with 0.5 and 1mm DRs respectively. Lag time of SMT permeation for untreated skin (passive permeation) was 3.56±1.06 hr and that attained after treatment of skin with 0.6, 0.9, 1.2 and 1.5 mm MNs were 2.69±0.24, 1.21±0.79, 1.09±0.25 and 0.24±0.14 hr respectively and 2.7±0.57 and 1.7±0.92 hr respectively with 0.5 and 1 mm DRs. Lag time values decreased with an increase in needle length.

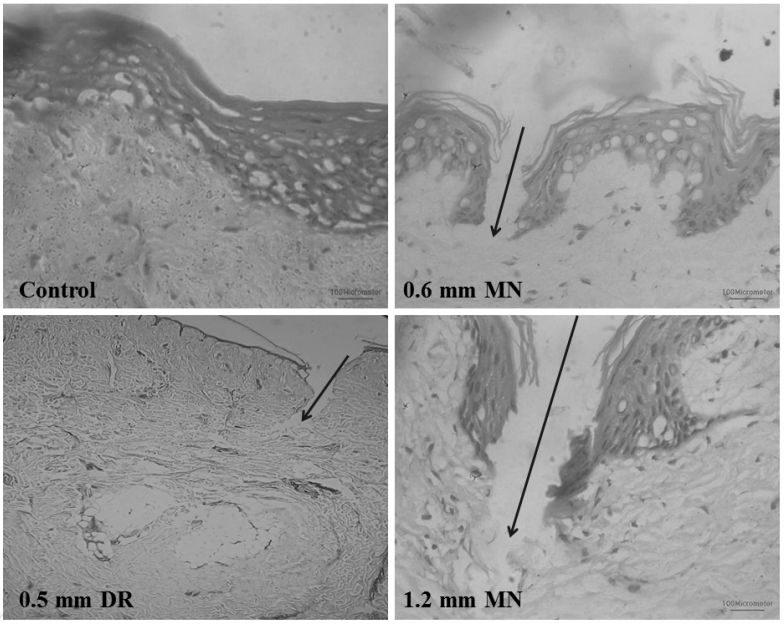

Fig. (4). Histological sections of untreated, 0.5mm DR, 0.6mm MN and 1.2mm MN treated skin samples.

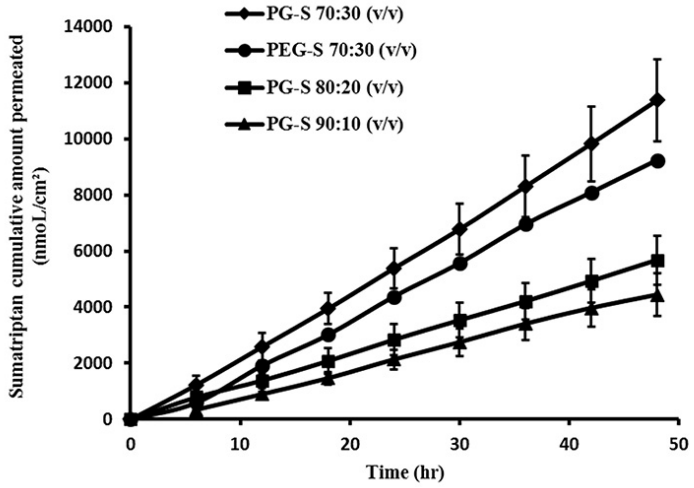

Fig. (5). Mean comparative *in vitro* skin permeation profiles of SMT obtained from PG-S combinations and PEG-S 70:30 (v/v).

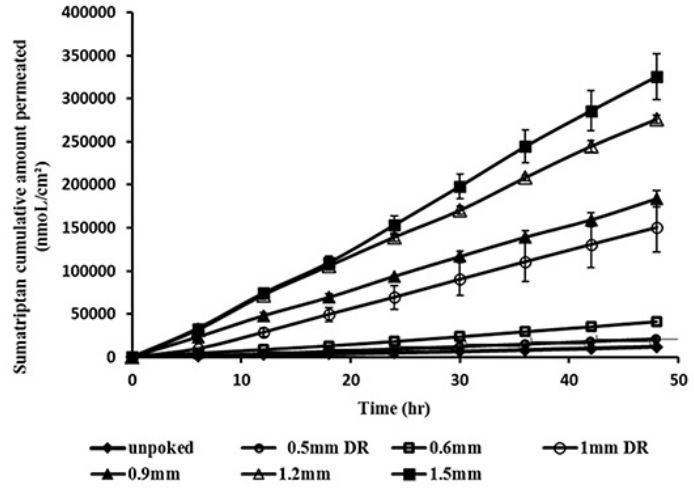

Fig. (6). Mean comparative *in vitro* skin permeation profiles of SMT obtained from passive and microneedle pretreatments.

Table 1. Permeation parameters of SMT with MN and DR in PG-S 70:30 (v/v) (Mean ± SD)*.

Skin treatment	Permeation Parameters		
	SMT Skin content (nmoL/gm)	Permeability coefficient (cm/hr)	Diffusion coefficient ($x10^{-04}$) (cm^2/sec)
Passive	9452.67 ± 698.33	1.69 ± 0.21	0.56 ± 0.07
0.5 mm DR	9476.57 ± 970.82	3.39 ± 0.62	1.12 ± 0.20
1 mm DR	27716.98 ± 11941.50	21.85 ± 3.29	7.28 ± 1.09
0.6 mm MN	17336.41 ± 2753.52	5.93 ± 0.55	1.98 ± 0.18
0.9 mm MN	42578.92 ± 4066.61	24.65 ± 2.29	8.22 ± 0.76
1.2 mm MN	51314.25 ± 696.72	37.30 ± 0.87	12.43 ± 0.29
1.5 mm MN	52410.75 ± 2461.28	44.70 ± 4.08	14.90 ± 1.36

*(n=3).

Enhancement ratios were also studied to evaluate the effect of the enhancement technique on the diffusion and permeation of SMT. Permeation parameters like diffusion coefficient and apparent permeability were computed from the obtained flux values of SMT (Table **1**).

DISCUSSION

The pig ear skin was preferred as a predictive paradigm in this study, for it is relatively thin and highly vascularized and is the best alternative to human skin [35-37]. Moreover, the lipid content of pig skin is close to that of human skin [38, 39].

Solubility Studies of SMT

Solubility studies of SMT performed with various combinations of 70:30, 80:20 and 90:10 %v/v of PG-S and PEG-S showed that SMT solubility has significantly increased (p <0.05) with increase in saline (S) percent in both vehicle systems, and was in the order of 70:30> 80:20> 90:10 %v/v (Fig. **3**). Furthermore, solubility of SMT was significantly higher in PG:S than in PEG:S at all the considered ratios ($p < 0.05$). Thus, PG:S vehicle combinations and also PEG:S at 70:30 %v/v were selected for further *in vitro* skin permeation studies.

Histological Examination of Skin Samples with and without Microneedle Treatment

The histological sections of the skin samples with and without microneedle treatment were prepared and observed (Fig. **4**) for the formation of microcondutis. From the micro photographs it is clearly evident that the microneedle treatment caused the disruption of stratum corneum and microneedles penetrated deeper into the tissue layers. It was also observed that larger and deeper microconduits were formed in skin as the microneedle length increased, and that MNs caused greater skin disruption than DRs, as determined from Toup View software. The histological evaluation of the skin sections suggested that the microneedles pierced the corneocytes and didn't merely indent them.

In-vitro Skin Permeation Studies

Passive Permeation

Passive permeation studies carried out using the selected donor vehicles indicated that SMT permeation was affected by the donor vehicle used and was found to be in agreement with solubility data. A 1.2 fold increase in the cumulative amount of SMT permeated was observed with PG:S at 70:30 %v/v as donor vehicle, when compared to PEG:S at 70:30 %v/v ($p < 0.05$) (Fig. **5**). This may be explained by the fact that PG can serve as a penetration enhancer leading to the extensive diffusion of drug molecules through the skin. PG can be used in intercellular diffusion enhancement by disordering the lamellar lipid structure. The lower penetration ability of PEG 400 may be because it is less effective in maintaining the skin surface as dehydrated skin; it is more resistant to penetration. PEG 400 was reported not only to alter skin barrier properties, but it may also form complexes with drugs in the receptor compartment and therefore retards diffusion [40, 41]. Since a significant difference was observed in the values obtained from PG:S and PEG:S combinations; PG:S at 70:30 %v/v was selected as donor vehicle for further microneedle assisted permeation studies.

Microneedle Treatment to the Skin

In the present study two commercially available stainless steel microneedle devices (Dermaroller® microneedle rollers and AdminPatch® microneedle arrays) of various needle lengths were exploited to overcome the lipophilic stratum corneum barrier. Microneedle treated permeation studies were carried out using PG:S at 70:30 %v/v as donor vehicle (Fig. **6**). Significant enhancement in permeation of SMT following microneedle pretreatment (both rollers and arrays) was observed when compared to that with microneedle untreated permeation studies ($p < 0.05$). A 3.11, 16.18, 24.25, 28.61; and 1.84, 13.22 fold increase in cumulative amount of SMT permeated was observed after treatment of skin with MNs (0.6, 0.9, 1.2, 1.5 mm lengths); and DRs (0.5, 1 mm lengths) respectively, and the flux values were increased by 3.50, 14.56, 22.03, 26.4; and 2.01, 12.91 orders of magnitude respectively when compared to passive SMT permeation data. Also, a 15.91 fold decrease in the lag time was ob-

served with 1.5 mm MN treatment when compared to passive permeation data. Both apparent permeability and diffusion co-efficient increased with the increase in needle length (Table **1**) with microneedle treatment. The values of these parameters were in agreement with the other permeation parameters results and showed a positive relationship between microneedle (length and type) and permeation enhancement of SMT. The results revealed that as the length of microneedles increased, the permeation of SMT increased significantly ($p < 0.05$); with about 28 fold increase in cumulative amount of SMT permeated with 1.5 mm MN treatment.

Moreover, the increase in SMT flux was only marginal (2-3 folds) when treated with shorter microneedle lengths (0.5, 0.6 mm), while that enormously increased on treatment with longer microneedles (12-26 folds). It was reported previously that the transepidermal water loss (TEWL), a parameter to estimate skin damage, remained at base levels after treatment of skin with shorter microneedles (0.6 mm) on the dermis side indicating that the skin layer disruption with shorter microneedles was minimal and in turn explaining the minimal enhancement in permeation observed in this study [42]. Overall, the enhancement in transdermal SMT permeation was in the order of 1.5 mm MN>1.2 mm MN>0.9 mm MN>1 mm DR>0.6 mm MN>0.5 mm DR> Passive permeation.

In this investigation, two techniques of microneedle application, i.e., arrays (MN) and rollers (DR), were employed with an aim to enhance transdermal SMT permeation. The data obtained with similar microneedle lengths (0.6 with 0.5 mm and 0.9 with 1 mm of MN and DR respectively) was compared to derive an understanding on their relative efficiency on such enhancement (Fig. **6**). From the cumulative amounts of SMT permeated at the end of 48 hrs, it was clearly evident that the increase in permeation on treatment with MN was considerably higher than that observed with DR of similar microneedle lengths.

The enhancement of transdermal permeation of drugs by disruption of skin integrity, as in the present study, is a complex phenomenon. The viscoelastic nature of skin makes it difficult to give an easy explanation or comparison of results obtained from different techniques of microneedle application. The interplay of many factors *viz.* microneedle density and distribution in a device, needle length and geometry, force of insertion, time of insertion, characteristics of the created microconduits and molecular properties of the drug, etc., determines the overall benefit of using any specific technique of microneedle application over others [31, 32]. However, the superior enhancement of SMT permeation with MN may be attributed majorly to two aspects or differences like force of insertion and density of microneedles. With regard to the force of microneedle application, the cylindrical assembly of microneedles on DR, a much lower force is employed during its application on the skin, as it only involves rolling of device over the skin surface [43]. However, manual application/pressing of MN exerts relatively higher force, which may have resulted in greater disruption of skin and responsible for such higher permeation. The depth of penetration obtained in our study with DR was found to be 30-40% while that with MN was 40-50% of microneedle length (as supported by histological studies), which further strengths the above statement regarding force of application and better barrier disruption with MN.

Furthermore, Dermarollers® possess 24 circular arrays of 8 needles each (total 192 needles) in a cylindrical assembly (with 2 cm diameter and 2 cm length of the cylinder); whereas, AdminPatch® arrays possess a number of 187, 85, 41 and 31 with an effective length of 0.5 mm, 0.8 mm, 1.1 mm and 1.4 mm respectively in a circular plate like structure of 1cm^2 area for 0.6, 0.9, 1.2 and 1.5 mm arrays. This higher density of microneedles in case of arrays when compared to rollers might also have contributed to the above observation. Also in general, the specific arrangement of MN favors more number of microneedles to penetrate through the skin compared to the DR arrangement of microneedles. The altered permeation enhancement by these microneedles might also be accredited to the differences in their geometries. Thus, it can be inferred that MNs may be beneficial over DRs in enhancing transdermal permeation of SMT. However, it may be stated that increase in microneedle length resulted in a significant increase in SMT permeation irrespective of the technique of microneedle application.

Assessment of SMT Flux Obtained to the Clinical Relevance

The surface area required for the transdermal systems (patch) to be effective for SMT delivery was calculated from the attained steady state flux values with passive and microneedle treatment permeation studies. From the literature, total body clearance (Cl_T) and steady-state concentration after a subcutaneous injection (C_{ss}) in humans are 70 L/h and 72 ng/mL respectively [12]. The predicted steady state plasma concentration of SMT following the application of the transdermal patch was calculated from the *in vitro* steady state flux by using the following equation:

$$C_{ss} = (J_{ss} .A)/Cl_T$$

Where, C_{ss} is the predicted steady state plasma concentration (ng/mL); J_{ss} is the steady state flux across human or pig ear skin; Cl_T is the total body clearance obtained after i.v administration in humans. Using the C_{ss}, Cl_T and J_{ss} values, the surface area required was calculated from the above equation.

The required surface for a transdermal system with the obtained passive flux was 66.69 cm^2 which may not be feasible in reality. However, after microneedle treatment, the necessary surface area to achieve the therapeutic significance was 19.04, 4.58, 3.03 and 2.53 cm^2 for 0.6, 0.9, 1.2 and 1.5 mm MNs respectively and 33.24 and 5.17 cm^2 respectively for 0.5 and 1mm DR. The results showed that with increase in needle length, the required surface area of transdermal system to deliver SMT at clinically relevant levels has significantly reduced. A maximum of 23 fold decrease in the required patch area was observed with 1.5 mm MN treatment of skin when compared to passive permeation, making transdermal delivery of SMT feasible under realistic conditions.

In summary, our results inferred that SMT permeation has significantly increased with the application of microneedles (more for MN than DR) when compared to passive diffusion and it was also dependent on the solubility of

SMT in the donor vehicle. Hence, microneedle assisted transdermal delivery of SMT can be seen as a good alternative route for its administration, with better patient compliance.

CONCLUSION

Transdermal drug delivery generally offers increased patient compliance owing to its ease of use. In case of chronic conditions like migraine, transdermal administration of drugs is more beneficial. The *in vitro* permeation studies of SMT demonstrated that microneedle pretreatment of skin can significantly enhance the SMT permeation when compared to passive permeation. Such enhancement was found to be dependent on the microneedle length and type of insertion, and was superior with application of microneedle arrays over rollers. The *in vitro* flux values also proved that microneedle treatment can enormously reduce the required surface area of a transdermal system when compared to that for untreated skin, making it very feasible for human use. Thus, SMT may be successfully delivered *via* transdermal route by microneedle application and it may be feasible to develop microneedle assisted transdermal delivery systems for SMT with clinically relevant levels of SMT.

CONFLICT OF INTEREST

The author(s) confirm that this article content has no conflict of interest.

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PATIENT CONSENT

Declared none.

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