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MECHANISMS OF TRANSDERMAL CONTROLLED NITROGLYCERIN ADMINISTRATION (I): DEVELOPMENT OF A FINITE-DOSING SKIN PERMEATION SYSTEM

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ABSTRACT

A new finite-dosing cell for in vitro skin permeation studies was recently developed to overcome the deficiencies observed in the commercially available Franz diffusion cell and to accomplish the solution hydrodynamics and temperature control required in studying the rate profiles of skin permeation.

Results of comparative studies indicated that the improved diffusion cell, named Keshary-Chien diffusion cell, can achieve and maintain the target body temperature on the skin surface and in the receptor solution, which cannot be accomplished by the Franz diffusion cell. Solution mixing efficiency was substantially improved, so the drug distribution and

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concentration homogeneity could be achieved in the Keshary-Chien diffusion cell within a duration four times shorter than in the Franz diffusion cell; and a 3-fold reduction in the thickness of the hydrodynamic boundary layer was achieved, so the effect of mass transfer in the hydrodynamic boundary layer on the skin permeation rate profiles was minimized.

The controlled release and skin permeation of nitroglycerin from four marketed transdermal therapeutic systems were investigated. As the result of the improvements in solution hydrodynamics and temperature control, the rates of release and skin permeation of nitroglycerin from the controlled-release transdermal therapeutic systems were enhanced substantially; so, the skin permeation rate profiles could be realized with minimal effect from the mass transfer process.

INTRODUCTION

Drug absorption by the skin can be measured directly in living humans or animals by analyzing the drug concentration profiles in the blood or in the urine following topical administration. However, a quantitative assessment of the mechanisms and rates of transdermal permeation of drug can be achieved by analyzing the drug permeation profiles through an excised skin mounted on a diffusion cell, without any unwanted complications from the specific pharmacokinetic rate processes, such as distribution, metabolism and excretion of a drug in the body.

The relevance of *in vitro* skin permeation studies using an excised skin has largely based upon the knowledge that the stratum corneum is the rate-limiting barrier to the skin permeation of drugs (1). In several studies, the percutaneous absorption data obtained by *in vitro* and *in vivo* techniques were compared and the results tended to support the contention that reliable measurements of transdermal bioavailability can be accomplished by *in*

vitro skin permeation studies using the excised skin (2-5). On the other hand, Wester and Maibach (6) reported that a more accurate assessment of skin permeability is obtained by employing the in vivo percutaneous absorption studies.

One of the most frequently used in vitro techniques for skin permeation studies is the finite-dosing technique developed by Franz (5). A diffusion cell, named Franz diffusion cell, was designed and applied, over the years, to study the percutaneous absorption of chemicals and drugs under a finite dose condition. A commercial model of the Franz diffusion cell has been marketed and used extensively for skin permeation studies (Figure 1).

Since the marketing introduction of several controlled release transdermal therapeutic systems, namely Transderm-Nitro and Transderm-Scop systems[2], Nitro-Dur system[3] and Nitrodisc system[4], a great extent of R & D interests in the transdermal controlled administration of a variety of drug entities have been triggered. To assist the development and the evaluation of a controlled-release transdermal therapeutic system, design of a sensitive, reproducible, and simple in vitro skin permeation cell is highly critical and in great demand. In view of these facts, the commercially available Franz diffusion cell was recently evaluated in this laboratory to assess its potential in studying the controlled release and skin permeation kinetics of nitroglycerin from three recently marketed one-a-day transdermal patches (7).

During the course of studying the mechanisms of transdermal controlled administration and of controlled release of drugs from the transdermal therapeutic systems, several deficiencies were noted in the Franz diffusion cell design: it could not achieve the solution hydrodynamics, mixing efficiency, and temperature control required in the quantitative evaluations

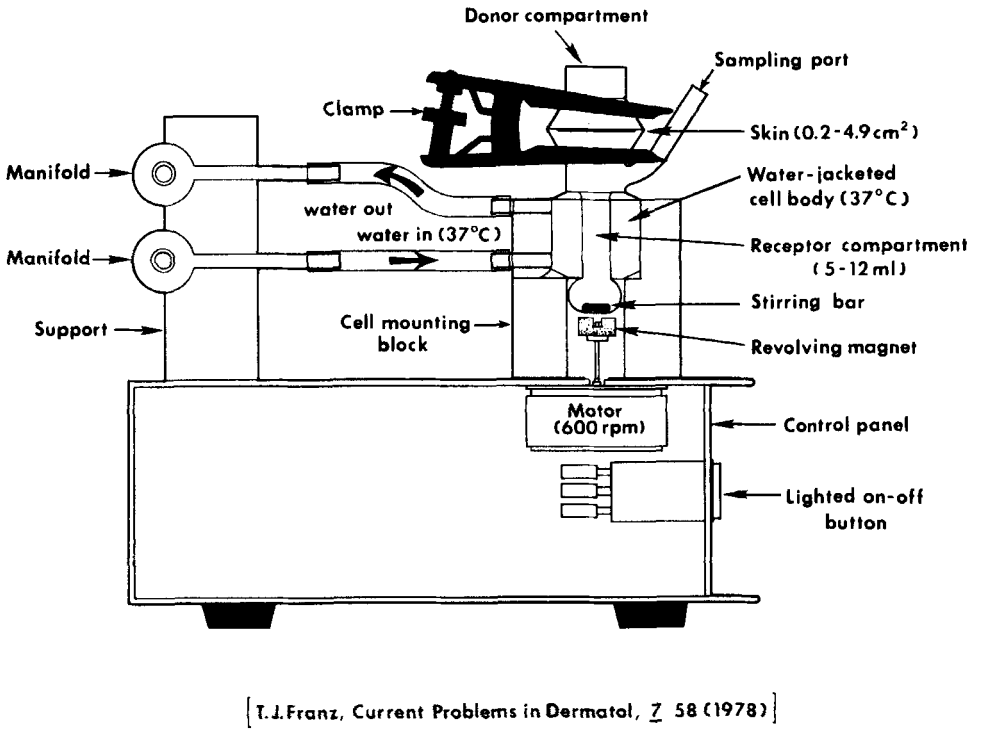

Figure 1: Schematic illustration of the commercially available finite-dosing Franz diffusion cell assembly. Each of the diffusion cells stations in a cell mounting block and consists of two compartments: a donor compartment, which is exposed to an ambient condition, and a receptor compartment, which is maintained at 37°C by circulating a thermostated water through the waterjacket. The solution hydrodynamics in the receptor compartment is kept at constant by a tiny rod-shaped magnet rotating at 600 rpm by a synchronous motor mounted underneath the cell mounting block.

of skin permeation kinetics. In view of this fact, a new finite-dosing skin permeation system was thus developed in this laboratory by improving the Franz diffusion cell design to overcome its inherent deficiencies outlined above. It has been designed in such a way that the new cell design can utilize the same driving unit and other accessories for the Franz diffusion cell by simple modification of the cell mounting block.

This newly improved skin permeation system was evaluated, in terms of solution hydrodynamics, mixing efficiency, temperature control as well as drug release and skin permeation kinetics, in comparison with the original Franz diffusion cell. Cell design and results of the evaluations will be discussed in the first report of this series of investigations.

EXPERIMENTAL

Materials:

1. Pure nitroglycerin - It was extracted from nitroglycerin-lactose triturate[5] (1:9) by dissolving the lactose base in excess amount of distilled water and then collecting the insoluble, pure nitroglycerin precipitate at the bottom of the glassware. After several washings, the purity of nitroglycerin was determined by the USP method[6] before uses.
2. Transdermal nitroglycerin patches - Four recently marketed transdermal therapeutic systems were evaluated as obtained in this investigation:
    1. Nitrodisc system[4] (8 cm^2; 16 mg)
    2. Nitro-Dur system[3] (10 cm^2; 51 mg)
    3. Transderm-Nitro system[2] (10 cm^2; 25 mg)
    4. Deponit system[7] (16 cm^2; 16 mg)
3. Reagents - Polythylene glycol 400[8], benzoic acid[8], sodium chloride[8], methanol[9] (Distilled-in-glass HPLC grade) were used as obtained. HPLC grade distilled water was prepared freshly before use[10].

Preparations:

1. Preparation of Benzoic Acid discs: A dual half-circular mold was used in the preparation. The mold was placed on a smooth pill tile, benzoic acid was fused and then poured into the mold. After solidifying, the disc was removed and reduced to a proper size by sandpapering it. In the present investigation, a disc of 18 mm in diameter and 5mm in thickness was mounted between the donor and the receptor compartments of the diffusion cells.
2. Preparation of skin: 5 to 7 weeks old male hairless mice[11] were sacrificed by dislocating the cervical part of the spinal cord. A piece of full-thickness skin sample (3.5 cm x 3.5 cm) was excised from the abdominal region (special care was taken not to damage the skin). The dermal sides of the skin sample was cleaned of any adhering subcutaneous tissue and/or blood vessels. The skin sample was then examined microscopically to make sure of the integrity of the skin.
3. Preparation of Elution Solution: It was freshly prepared everyday in normal saline to contain 20% w/w of polyethylene glycol 400.

Analytical methods:

1. Spectophotometric analysis of benzoic acid - A microprocessor-controlled UV/Vis spectrophotometer[12] was used. Spectrogram was scanned for each sample solution, after proper dilution, from 240 to 210 nm; A characteristic peak was observed for benzoic acid at a λmax of 222 nm. The absorbance value at the peak was read and the correspondent benzoic acid concentration was calculated from the calibration curve established by reference standard solution.
2. HPLC analysis of nitroglycerin - For this investigation, a

microprocessor-controlled high performance liquid chromatograph[13] equipped with a variable wavelength detector, an automatic sampler, a variable-volume injector, a dual-head reciprocating pump and a dual solvent system was used. Using a combination of methanol and distilled water (6:4) as the mobile phase at a flow rate of 1 ml/min. and the column temperature at ambient, nitroglycerin in the sample solution, (with injection volume of 10 µl) was resolved by a reversed phase column and detected at a wavelenth of 205 nm.

Under the HPLC conditions outlined above, nitroglycerin produced a very sharp, clear absorption peak at a retention time of 4.9 min, while the two primary degradation products, 1,2- and 1,3-dinitroglycerin, had yielded characteristic peaks at retention time of 3.1 and 2.9 min., respectively. This stability-indicating method has a detection sensitivity of 75 - 100 ng/ml for nitroglycerin.

Design of an Improved Finite-dosing Diffusion Cell:

A unit of the currently marketed Franz diffusion cell is depicted in Figure 2 with the improved finite-dosing diffusion cell developed in this investigation. To enhance the mixing efficiency, the inner diameter of the receptor solution compartment was widened from 12.5 mm to 20 mm, while the height was reduced from 68.5 mm to 50 mm. In addition to the change in the dimension, the stirring bar was replaced with a star-head magnet to improve the flow pattern in the receptor solution. In the meantime, the water-jacket compartment was extended to envelope a greater surface area of the receptor compartment than the Franz diffusion cell to provide a better temperature control and equilibrium. The sampling port in the improved diffusion cell was modified to permit stoppering with

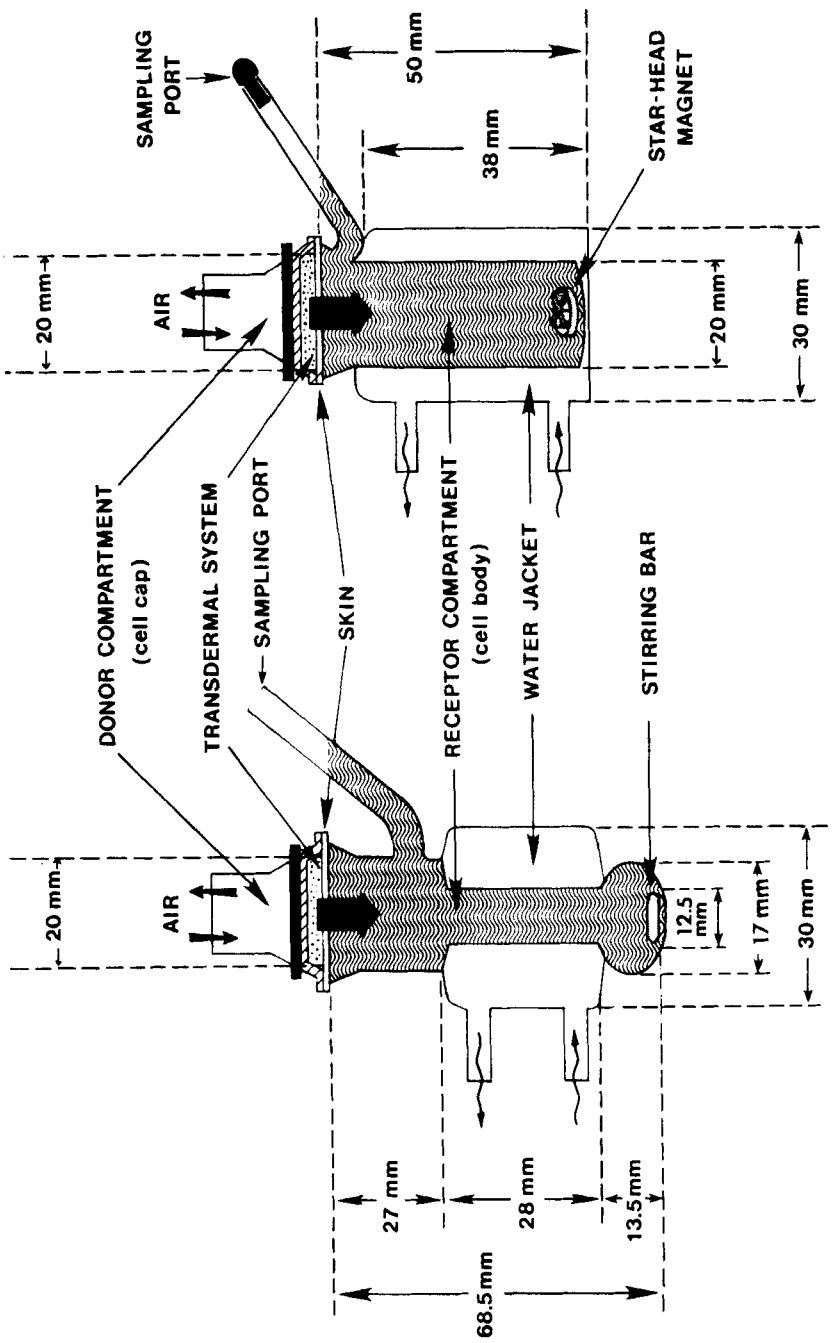

Figure 2: Diagrammatic illustration and comparison of Franz and Keshary-Chien diffusion cells.

a matched glass stopper to eliminate any possible evaporation of the elution solution from the receptor compartment.

The changes outlined above were made in such a way that the new cell design can utilize the same driving unit and other accessories for the original Franz diffusion cell after a minor modification of the cell mounting block.

This improved finite-dosing diffusion cell, which is designated as the Keshary-Chien diffusion cell for the purpose of easy identification, was calibrated in four areas: (a) thickness of hydrodynamic boundary layer, (b) dynamics of solution temperature, (c) dynamics of skin temperature, and (d) efficiency of solution mixing. The procedures for calibration are outlined as follows:

a) Determination of Hydrodynamic Boundary Layer Thickness - A benzoic acid disc (18 mm in diameter; 5mm in thickness) was prepared using the method described earlier (8). It was mounted between the donor and receptor compartments of the Franz diffusion cell and the Keshary-Chien diffusion cell and the receptor compartment was then filled with the saline solution containing various volume fractions of PEG 400 to conduct the dissolution study of the benzoic acid disc. At a predetermined time interval, sample was withdrawn from the receptor solution and assayed for benzoic acid concentration, after appropriate dilution, by the spectrophotometric method described earlier. The receptor compartment was refilled with the same volume of drug-free saline/PEG 400 solution immediately after sample withdrawal.

b) Determination of Solution Temperature Profiles - While maintaining the receptor compartment of both diffusion cells at 37°C by a circulating waterbath[15], a saline solution at 21°C was filled into the receptor

compartment to the rim, which was exposed to the atmosphere at 25°C. The solution temperature at the upper surface, which was expected to be in intimate contact with the dermal surface after a skin sample was mounted onto the diffusion cells, was monitored by a sensitive temperature probe[16] as a function of time until an equilibrium temperature was reached and maintained at steady state for at least 30 minutes. At the steady state, the solution temperatures at the mid-point and the bottom of the receptor solution were also recorded for comparison.

c) Determination of Skin Temperature Profiles - An adequate size of male, hairless mouse abdominal skin was freshly excised and mounted onto the Franz and the Keshary-Chien diffusion cells (Figure 2) with stratum corneum facing up into the donor compartment, which was exposed to the atmosphere at 25°C, and the dermis facing down into the receptor compartment, which was maintained at 37°C by a circulating waterbath[15]. A saline solution at 20.9°C was filled into the receptor compartment to the level in a close contact with the dermal surface. The skin temperature on the stratum corneum surface was then monitored as a function of time also by the temperature probe[16] until an equilibrium temperature was reached and maintained at steady state for at least 30 minutes.

d) Determination of Solution Mixing Efficiency - After completion of the solution temperature profile studies outlined above, the solution at the equilibrium temperature was spiked with a known quantity of nitroglycerin from the top of the receptor solution and samples were taken at predetermined time intervals from the bottom of the receptor

solution with the magnet rotating at 600 rpm. The samples were assayed for nitroglycerin concentration by the HPLC method described earlier. Cares were taken in the spiking and in the withdrawal of the receptor solution to avoid any turbulent effect to the solution.

e) Determination of Drug Release Profiles - The transdermal therapeutic system was clamped between the donor and the receptor compartments of the Franz and Keshary-Chien diffusion cells, with the drug releasing surface facing into the receptor compartment, which was maintained at 37°C by a circulating waterbath[15]. Receptor compartment was filled with an elution solution containing 20% w/w PEG 400 in normal saline. At a predetermined time interval, the elution solution was completely emptied and was then filled with the equal volume of fresh (drug-free) elution solution. Samples were analyzed for nitroglycerin concentration by the HPLC method outlined earlier.

f) Determination of Skin Permeation Profiles - Skin sample (3.5cm x 3.5cm) was freshly excised from the abdominal region of the hairless mouse (male, 5-7 weeks old) and mounted onto the Franz and the Keshary-Chien diffusion cells, with the stratum corneum facing up into the donor compartment. The receptor compartment was maintained at 37°C by a circulating waterbath[15]. A unit of transdermal therapeutic system was placed over the skin with the drug-releasing surface in intimate contact with the stratum corneum and the whole assembly was clamped together with the donor cap on the top (Figure 2). The receptor compartment was then filled with the elution solution (preheated to 37°C). At predetermined time intervals, samples of the elution solution in the receptor compartment were withdrawn and replaced with fresh

(drug-free) elution solution. Concentration of nitroglycerin in the samples was determined by the HPLC method outlined earlier.

RESULTS AND DISCUSSION

A unit of the improved finite-dosing diffusion cell, named Keshary-Chien diffusion cell, developed in this laboratory is illustrated side-by-side with a unit of the commercially available Franz diffusion cell in Figure 2. To improve the temperature control on the skin surface and in the receptor solution as well as to enhance the efficiency of solution mixing and the distribution of drug solute following skin permeation, several improvements were made: (a) The inner diameter of the receptor solution compartment was widened from 12.5 mm to 20 mm, while the height was reduced from 68.5 mm to 50 mm; (b) The rod-shaped stirring bar was substituted with a more effective starhead-type magnet to improve the flow pattern of solution in the receptor compartment; (c) The water-jacket compartment was extended from 28 mm to 38 mm in height to cover a greater surface area of the receptor compartment to provide a better temperature control; and (d) The sampling port was modified, so it can now be totally enclosed with a matched glass stopper to eliminate any potential evaporation of the elution solution from the receptor compartment. These improvements were completed aiming to overcome the deficiencies noted in the Franz diffusion cell, while the external features of the Franz diffusion cell was still retained; so the Keshary-Chien diffusion cell can utilize the same driving unit and other accessories originally designed for the use of Franz diffusion cell.

The Keshary-Chien diffusion cell was calibrated, using the current marketed version of Franz diffusion cell as the reference, in the following

areas:

Thickness of Hydrodynamic Boundary Layer

When a fluid flows through a surface under conditions in which turbulence generally prevails, there is a thin film of fluid exists in laminar flow immediately adjacent to the surface. This is followed by a transition zone when the flow gradually changes to the turbulent flow existing in the outer region of the fluid. The rate of mass transfer of a drug solute through these three regions will depend upon the nature of the fluid motion prevailing in each region (9).

To examine the difference in the hydrodynamic condition for the fluid flow in the receptor solution and the effective thickness of the hydrodynamic boundary layer on the surface of the dermis between Keshary-Chien and Franz diffusion cells, the dissolution of benzoic acid disc in the saline solution containing various volume fractions of PEG 400, used as the solubilizer, was studied. The rate of dissolution of benzoic acid disc can be described by the following relationship:

$$\frac{dC}{dt} = \frac{k_m A}{V} (C_s - C_t) \qquad \text{(Eq. 1)}$$

where k_m is the mass transfer coefficient; A is the effective surface area of the disc; V is the volume of the dissolution medium; C_s is the saturation solubility of benzoic acid in the dissolution medium containing various volume fractions of PEG 400; and, C_t is the concentration of benzoic acid in the dissolution medium at time t.

Integration of Equation (1), under the initial condition of $C_t = 0$ at $t = 0$, gives:

$$\ln\left(\frac{C_s}{C_s - C_t}\right) = k_m \frac{A}{V} t \qquad \text{(Eq. 2)}$$

A linear relationship should be obtained when the dissolution data are plotted as $\ln (C_s/C_s - C_t)$ vs. (A/V)t (Figure 3). From the slope, the mass transfer coefficient (k_m) can be calculated. Results indicated that the dissolution profile of benzoic acid disc in the Keshary-Chien diffusion cell produces a higher k_m value than in the Franz diffusion cell.

Theoretically, k_m is related to three dimensionless numbers: Sherwood (Sh), Modified Reynold ($\bar{R}e$) and Schmidt (Sc) numbers by the following equation:

$$k_m = \frac{Sh.D}{d} = \frac{Const.D}{d} \bar{R}e^m \, Sc^{1/3} \qquad \text{(Eq. 3)}$$

where D = Effective diffusivity in a dissolution medium.

d = Diameter of the dissoluting surface of the benzoic acid disc.

m = Exponent; 0.7966 for Franz diffusion cell and 0.9834 for Keshary-Chien diffusion cell.

On the other hand, the thickness of the hydrodynamic boundary layer, h_d, is related to k_m by the following relationship:

$$h_d = \frac{D}{k_m} \qquad \text{(Eq. 4)}$$

Using Equation (3), the mass transfer coefficient (k_m) for a drug solute can be determined if D is known or predetermined. Similarly, the thickness of hydrodynamic boundary layer (h_d) can be calculated from Equation (4) if D and k_m are available. Alternatively, the effective diffusivity (D) of

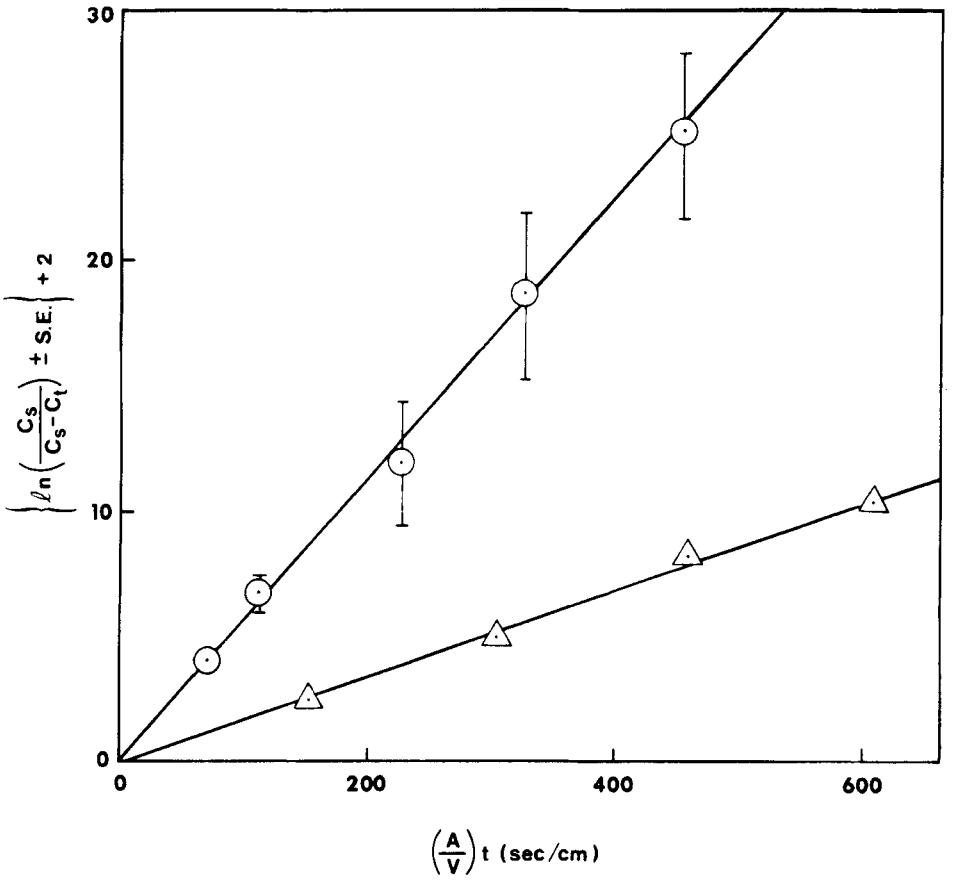

Figure 3: Linear relationship between ln (C_s/C_s-C_t) and (A/V)t for the dissolution of benzoic acid disc in the saline solution containing 20% $^{w}/_{w}$ PEG 400 as defined by Equation (2). Keys: ○ Keshary-Chien diffusion cell (k_m = 5.42 x 10^{-4} cm/sec), △ Franz diffusion cell (k_m = 1.63 x 10^{-4} cm/sec). Each of the data points represents the mean ± S.EM. of 3-6 determinations.

a drug solute can be computed from the diffusivity of another drug solute by the following equation (10):

$$D_n = \left(\frac{M_b}{M_n}\right)^{\frac{1}{2}} D_b \qquad \text{(Eq. 5)}$$

Where D_n and D_b are the effective diffusivities and M_b and M_n are the molecular weights for drug solutes, n and b, respectively.

Using Equations (3) - (5), the k_m and h_d values in Franz and Keshary-Chien diffusion cells were determined. Results (Table 1) indicated that the thickness of hydrodynamic boundary layer existing in the Keshary-Chien diffusion cell for the diffusion of benzoic acid and nitroglycerin (101 and 108 microns, respectively) is three folds thinner than that in the Franz diffusion cell (337 and 304 microns, respectively). Considering the thickness of hairless mouse skin, i.e., 379 microns (11), the effect of mass transfer process in the hydrodynamic boundary layer could have a greater contribution to the overall rates of release and skin permeation of benzoic acid and nitroglycerin in the Franz diffusion cell, due to its thicker hydrodynamic boundary diffusion layer, than in the Keshary-Chien diffusion cell.

Solution Temperature Profiles

The time course for the solution temperature in the receptor compartment to reach an equilibrium temperature is illustrated in Figure 4. Results indicated that as the receptor compartment is maintained at 37°C and the donor compartment is exposed to the atmosphere controlled at 25°C, it takes 10 minutes for the saline solution in the Keshary-Chien cell to reach an equilibrium temperature of 36.7°C on the solution surface, i.e., at the interface of donor and receptor compartments, from a starting

TABLE 1

Comparison in Mass Transfer Coefficient (k_m) and Hydrodynamic Boundary Layer Thickness (h_d) between Franz and Keshary-Chien Diffusion Cells

Diffusion Cells	k_m [1] ($^{cm}/_{sec} \times 10^4 \pm SE$)		h_d [2] ($cm \times 10^4 \pm SE$)	
	Benzoic Acid	Nitroglycerin	Benzoic Acid	Nitroglycerin
Franz	1.63 ± 0.12 [3]	1.32 ± 0.10	337 ± 25	304 ± 20
Keshary-Chien	5.42 ± 0.76 [4]	3.73 ± 0.52	101 ± 14	108 ± 15

1) Calculated from Equation (3)

2) Calculated from Equation (4)

3) Mean (± standard error) of 6 determinations

4) Mean (± standard error) of 3 determinations

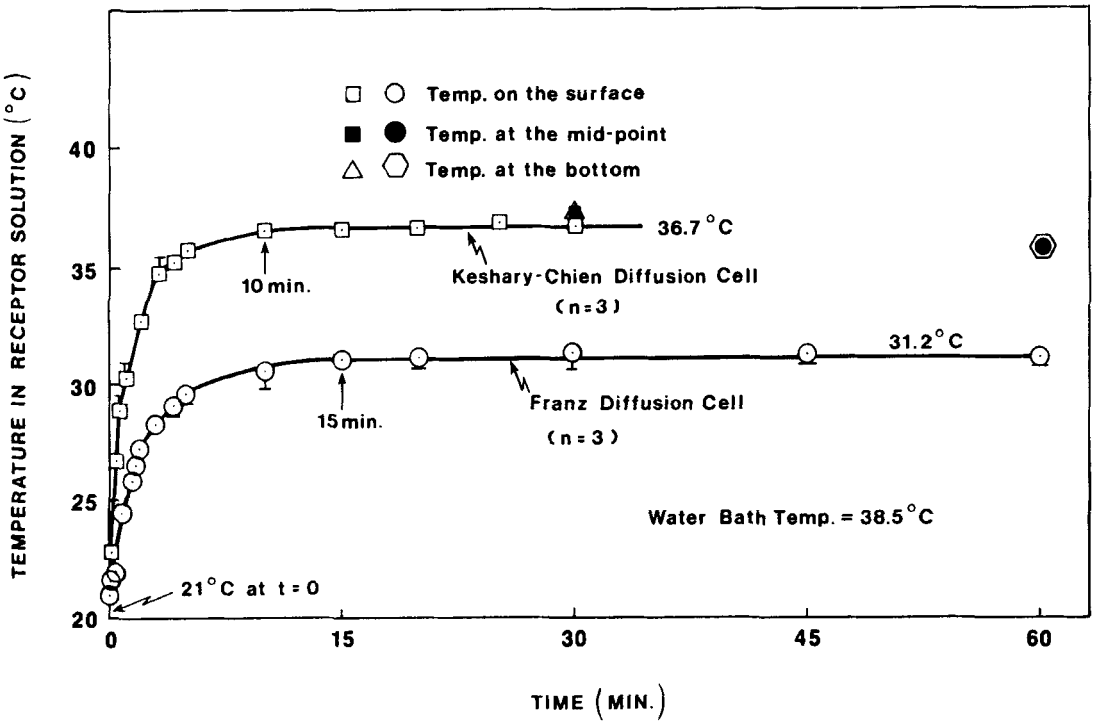

Figure 4: Time course for the solution temperature in the receptor compartment to reach an equilibrium temperature. Saline solution at 21°C was filled into the receptor compartment maintained at 37°C by a circulating waterbath, while the upper surface was exposed to the room temperatures at 25°C, and the temperature was monitored by a temperature probe as a function of time. In the Keshary-Chien diffusion cell, an equilibrium temperature of 36.7°C was reached within 10 minutes. On the other hand, in the Franz diffusion cell it took 15 minutes to reach a lower equilibrium temperature of 31.2°C.

temperature of 21°C. This equilibrium temperature stayed at steady state throughout the course of experiment, while the solution temperature at the mid-point and at the bottom of receptor compartment achieved the target temperature of 37°C. The differential solution temperature gradient was only 0.3°C throughout the receptor solution compartment.

On the other hand, it took 15 minutes for the saline solution in the Franz diffusion cell, under the identical experimental conditions, to reach an equilibrium temperature of 31.2°C on the solution surface, while the solution temperature at the mid-point and at the bottom of receptor compartment achieved an equilibrium temperature of 36.1°C. The equilibrium surface temperature (31.2°C) in the region, where the receptor solution was expected to be in intimate contact with the dermal surface after a skin sample was mounted between the donor and the receptor compartments, was found to be 5.8°C lower than the target temperature of 37°C as compared to a differential temperature of only 0.3°C in the Keshary-Chien diffusion cell. As mass transfer in the solution medium is an energy-dependent process, the greater differential temperature gradient (5.8°C) observed in the Franz diffusion cell could have a significant effect on the rate of mass transfer and, hence, the rates of release and skin permeation of drug from a drug delivery system.

The effect of temperature difference between Keshary-Chien and Franz diffusion cells on the rate of nitroglycerin release was evaluated in the Keshary-Chien cell. The rate of release of nitroglycerin from Nitro-Dur system was observed to decrease from 1,313.7 $mcg/cm^2/hr^{½}$ to 1,150.8 $mcg/cm^2/hr^{½}$ when the solution temperature immediately underneath the drug delivery system was reduced from 36.7°C to 31.2°C, which is equal

to a reduction of 12.4% in the release rate of nitroglycerin when the temperature was reduced by 5.5°C. Therefore, the contribution to the drug release rate profiles due to the temperature difference could account for only 12.4%. The remaining 87.6% of contribution could be attributed to other improvements realized in the Keshary-Chien diffusion cell, such as the reduction in the thickness of hydrodynamic diffusion layer.

Skin Temperature Profiles

The time course for the temperature on the skin surface to reach an equilibrium temperature is illustrated in Figure 5. Results indicated that as the receptor compartment is maintained at 37°C and the donor compartment is exposed to the atmosphere controlled at 25°C, it takes 15 minutes for the Keshary-Chien diffusion cell to reach an equilibrium skin surface temperature of 36.8°C, which is only 0.2°C lower than the target temperature of 37°C. On the other hand, it took 20 minutes to reach an equilibrium temperature of 33.2°C, which was 3.8°C lower than the target temperature.

In view of the fact that the skin permeation of drugs is an energy-dependent process (8), the 3.6°C-difference in the skin surface temperature between Franz and Keshary-Chien diffusion cells (33.2°C vs. 36.8°C) could influence the rate of skin permeation of drugs.

Efficiency of Solution Mixing

The efficiency of solution mixing was investigated by studying the time course required for a homogeneous distribution of nitroglycerin in the receptor solution following drug instillation (Figure 6). Results indicated that it takes only 2.5 minutes for the Keshary-Chien diffusion cell to achieve a homogeneous drug distribution, as indicated by the achievement of the

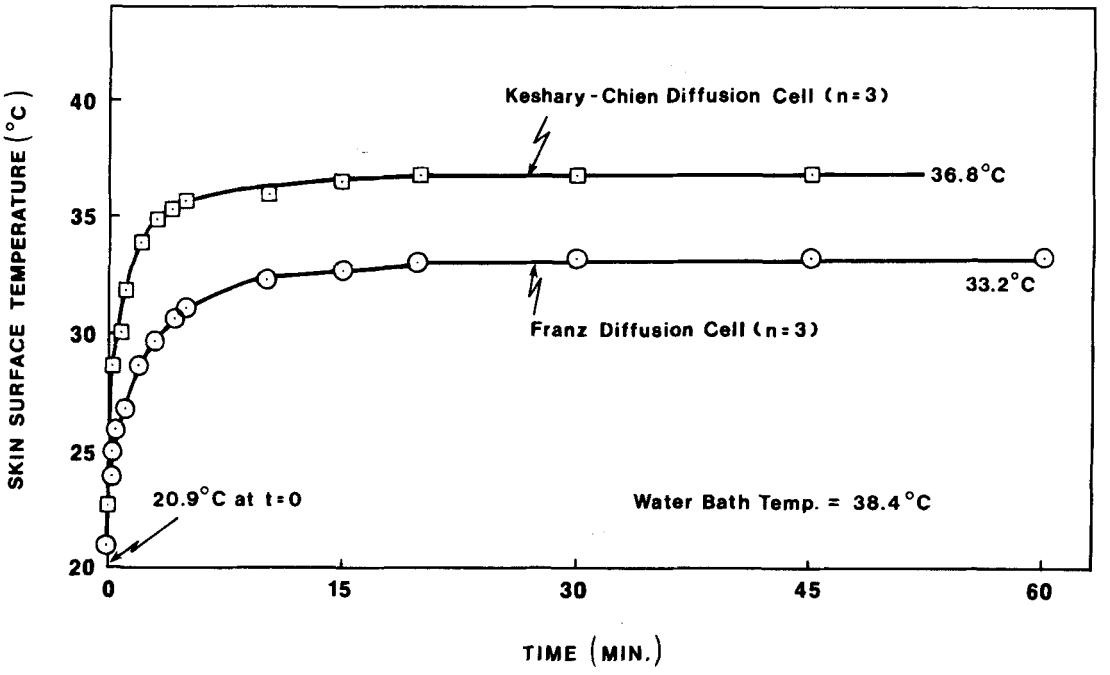

Figure 5: Time course for the temperature on the skin surface to reach an equilibrium temperature. Saline solution at 20.9°C was filled into the receptor compartment maintained at 37°C by a circulating waterbath, while the skin surface in the donor compartment was exposed to the room temperature at 25°C, and the skin surface temperature was monitored by a temperature probe as a function of time. For the Keshary-Chien diffusion cell, an equilibrium temperature of 36.8°C was reached within 15 minutes. For the Franz diffusion cell, an equilibrium temperature of 33.2°C was reached within 20 minutes.

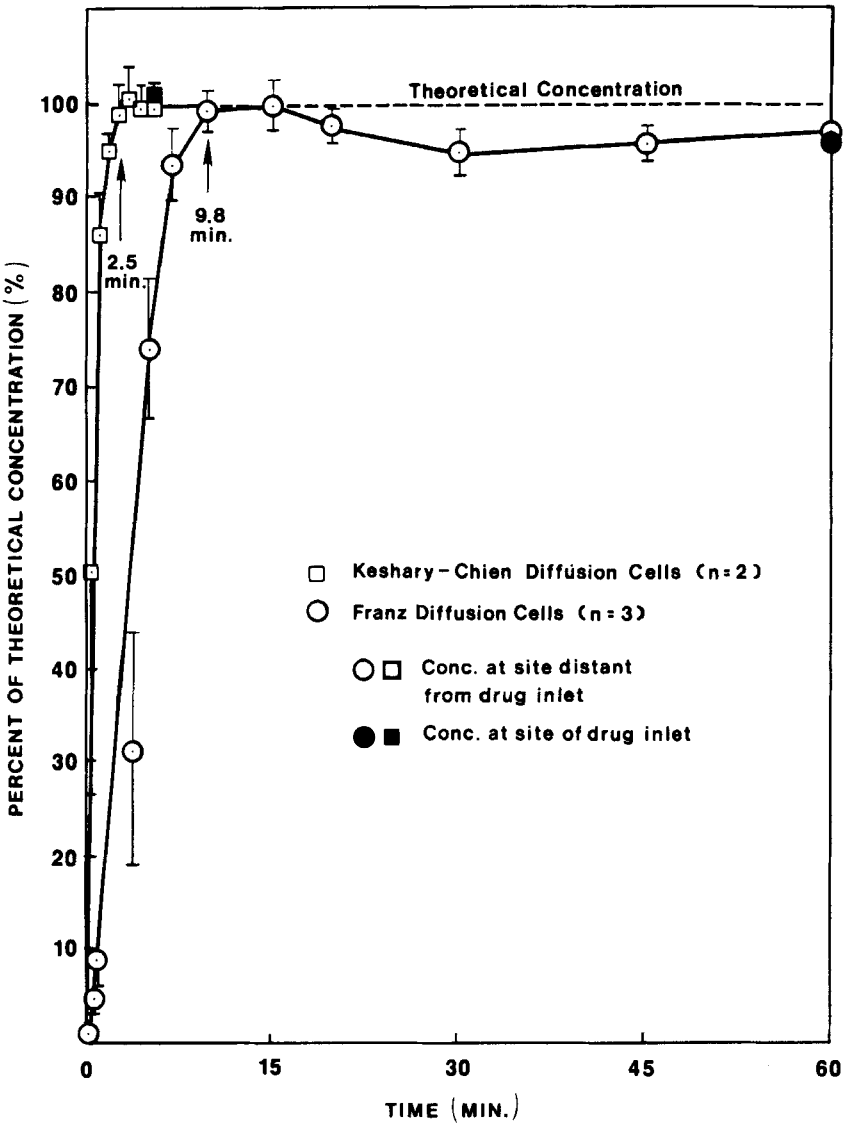

Figure 6: Time course for the concentration of nitroglycerin in the receptor solution to reach a theoretical level. It took only 2.5 minutes for the Keshary-Chien diffusion cell as compared to 9.8 minutes for the Franz diffusion cell.

100% theoretical drug concentration level throughout the receptor solution, as compared to 9.8 minutes for the Franz diffusion cell.

The results of hydrodynamic boundary layer thickness determination, receptor solution and skin surface temperature measurements, and solution mixing efficiency studies all demonstrated that the improvements made in the Keshary-Chien diffusion cell have overcome the deficiencies noted in the Franz diffusion cell and resulted in a substantial improvement of the temperature control on the skin surface and in the receptor solution, a significant enhancement of the efficiency of solution mixing and drug distribution, as well as a great reduction of the thickness of hydrodynamic boundary layer on the dermal surface. These improvements should increase the rates of drug release and of skin permeation; therefore, the mechanisms of controlled drug release and the fundamentals of skin permeation can be investigated.

Drug Release and Skin Permeation Kinetics

The improvements made in the Keshary-Chien diffusion cell over the Franz diffusion cell in the efficiency of solution mixing, thickness of hydrodynamic boundary layer and the control of skin surface and solution temperatures were observed to produce higher rates of controlled release and skin permeation of nitroglycerin from Nitro-Dur system (Figure 7), a matrix diffusion-type transdermal therapeutic system, and from Transderm-Nitro system (Figure 8), a membrane permeation-type transdermal therapeutic system.

The release of nitroglycerin from the membrane permeation-controlled Transderm-Nitro system was increased by 270% from 35.1 ± 2.9 mcg/cm^2/hr (in Franz diffusion cell) to 94.6 ± 4.1 mcg/cm^2/hr (in Keshary-Chien diffu-

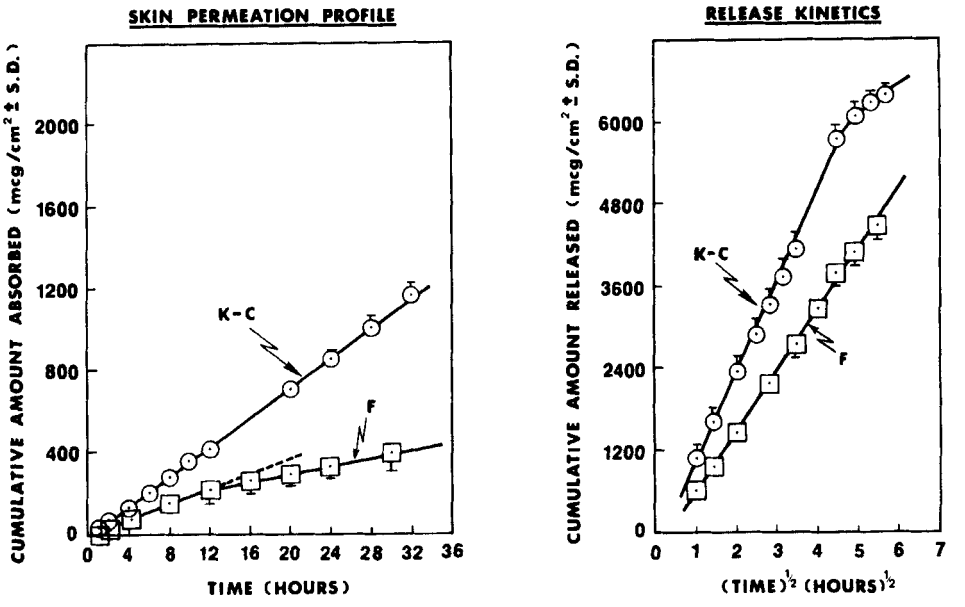

Figure 7: Comparison in the skin permeation profiles of nitroglycerin, thru hairless mouse skin, and the release profile from the matrix diffusion-controlled Nitro-Dur system between the Keshary-Chien and Franz diffusion cell designs. Greater rates of skin permeation and release of nitroglycerin were yielded with the Keshary-Chien (K-C) system than with the Franz (F) cell.

sion cell) (Table 2), while the mechanism of release studied by both diffusion cells followed the same zeroth order rate profile (Figure 8). The release of nitroglycerin from the Deponit system was also observed to give a constant, zeroth order rate profile as the Transderm-Nitro system with the rate of release increased by 133% from 13.5 ± 0.9 mcg/cm^2/hr (in Franz diffusion cell) to 18.0 ± 1.0 mcg/cm^2/hr (in Keshary-Chien diffusion cell). Furthermore, it was noted that the rate of skin permeation of nitroglycerin from Transderm-Nitro system is enhanced by 167% (from 14.1 ± 2.5

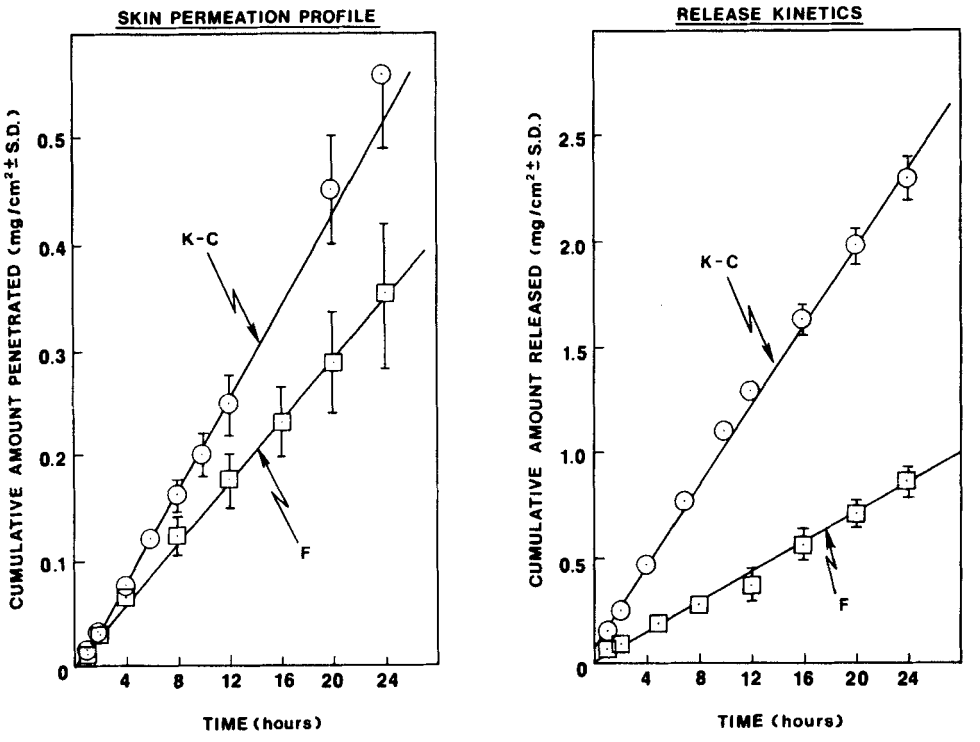

Figure 8: Comparison in the skin permeation profiles of nitroglycerin, thru the hairless mouse skin, and the release profile from the membrane permeation-controlled Transderm-Nitro system between the Keshary-Chien and Franz diffusion cell designs. Greater rates of skin permeation and release of nitroglycerin were produced with the Keshary-Chien (K-C) system than with the Franz (F) cell.

mcg/cm^2/hr to 23.6 ± 2.9 mcg/cm^2/hr), while an increase of 153% is noted in the Deponit system (Table 3). Deponit system is a multilaminate system with nitroglycerin depot covered by an adhesive layer.

The release of nitroglycerin from the matrix diffusion-controlled Nitro-Dur system was observed to follow a linear $\underline{Q \text{ vs. } t^{1/2}}$ relationship

TABLE 2

Comparison in Rate of Release of Nitroglycerin from Various Marketed Transdermal Therapeutic Systems

Systems	Release Rate	
	Franz Diffusion Cell	Keshary-Chien Cell
Transderm-Nitro ($mcg/cm^2/hr$)	35.1 ± 2.9	94.6 ± 4.1
Deponit ($mcg/cm^2/hr$)	13.5 ± 0.9	18.0 ± 1.0
Nitro-Dur ($mcg/cm^2/hr^{½}$)	841.6 ± 19.1	1,303.2 ± 18.7
Nitrodisc ($mcg/cm^2/hr^{½}$)	498.5 ± 27.7	552.4 ± 51.4

TABLE 3

Comparison in Rate of Skin Permeation of Nitroglycerin from Various Marketed Transdermal Therapeutic Systems

Systems	Skin Permeation Rate ($mcg/cm^2/hr$)	
	Franz Diffusion Cell	Keshary-Chien Cell
Pure Nitroglycerin	19.8 ± 3.4	41.4 ± 4.9
Deponit	7.3 ± 1.3	11.2 ± 1.8
Transderm-Nitro	14.1 ± 2.5	23.6 ± 2.9
Nitro-Dur	17.0 ± 2.8	31.1 ± 1.9
Nitrodisc	17.7 ± 2.8	44.7 ± 6.4

when evaluated by both diffusion cells (Figure 7). The rate of release was found to increase by 155% (from 841.6 ± 19.6 mcg/cm^2/hr^{½} in Franz diffusion cell to 1,303.2 ± 18.7 mcg/cm^2/hr^{½} in Keshary-Chien diffusion cell) (Figure 7 and Table 2). Similarly, the rate of release of nitroglycerin from the Nitrodisc system was enhanced by 111% from 498.5 ± 27.7 to 552.4 ± 51.4 mcg/cm^2/hr^{½} (Table 2). Nitrodisc system is a microsealed drug delivery system (12-17).

On the other hand, the skin permeation of nitroglycerin released from Nitro-Dur and Nitrodisc systems was observed to follow a linear Q vs. t relationship (Figure 7). The rate of skin permeation was increased by 183% for Nitro-Dur system and by 253% for Nitrodisc system (Table 3), when the Keshary-Chien diffusion cell was used to replace the Franz diffusion cell to study the kinetics of skin permeation. It was also interesting to note that a perfect Q vs. t linearity was obtained for the skin permeation of Nitro-Dur system when studied by Keshary-Chien cell (Figure 7).

Even the rate of skin permeation of pure nitroglycerin, without any influence from the controlled release mechanism of the transdermal therapeutic system, was also observed to increase by 209% when studies were carried out in Keshary-Chien diffusion cell than in the Franz diffusion cell.

The increased rates of release and skin permeation of nitroglycerin observed, with and without the modulation of controlled-release transdermal therapeutic systems, were apparently the results of the improvements in the efficiency of solution mixing, which produces a smaller thickness of hydrodynamic boundary layer and a better homogeneity of drug concentration in the solution, as well as in the temperature control of skin

surface and receptor solution, which has a minimum temperature variation along the diffusion path for the skin permeation of nitroglycerin.

It was reported earlier that there is a temperature difference of 5.5°C in the receptor solution between Keshary-Chien and Franz diffusion cells in the region immediately underneath the dermal surface (Figure 4). This differential temperature yielded a difference of 12.4% in the release rate of nitroglycerin from Nitro-Dur system. The observation implied that another 87.6% of the difference in the rate of release could be attributed to the effect of mass transfer in the hydrodynamic diffusion layer, which has a three-fold difference in thickness between Keshary-Chien and Franz diffusion cells.

In other words, the increased rates of controlled release and skin permeation of nitroglycerin observed in the Keshary-Chien diffusion cell could be attributed to the combining effect of the reduced thickness of hydrodynamic boundary layer and the better control of temperature in the diffusion path. The contribution from the hydrodynamic boundary layer could constitute the principal one.

FOOTNOTES

1. Crown Glass Company, Somerville, New Jersey
2. Ciba Pharmaceutical Corporation, Summit, New Jersey
3. Key Pharmaceuticals, Inc., Miami, Florida
4. Searle Pharmaceutical Corporation, Skokie, Illinois
5. Imperial Chemical Company, Willmington, Delaware
6. USPXX, page 552
7. Pharma-Schwarz GmHb, Monheim, Germany
8. Fisher Scientific Co., Fairlawn, New Jersey

9. Burdick & Jackson Lab., Inc. Muskagen, Michigan
10. By "Nanopure" Sybron/Barnstead, Boston, Massachusetts
11. HRS/J Strain, Jackson Laboratories, Bar Harbor, Maine
12. UV/Vis Spectrophotometer, Model 559A, Perkin-Elmer Corp., Oak Brook, Illinois
13. HP Model 1084B HPLC, Hewlett-Packard, Palo Alto, California
14. HP, RP-8 (10 μm, 200 x 4.6 mm), Hewlett-Packard, Palo Alto, California
15. Circulator, Model 80, Fisher Scientific Company, Fairlawn, New Jersey
16. Thermialert, Model TH-6D, Bailey Instruments Inc., Saddle Brook, New Jersey

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